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ERRATA.

Page 11, Heading of col. 1, for " Case No." read " Type of Locality "
Page 76, col. 1, line 29, for " *Gonombombyx* " read " *Gonobombyx* "
Page 79, col. 1, 5 lines from end, for " *rubricinctus* " read " *rubrocinctus* "
Page 110, Table VII, col. 4 under Turabi, for " +·12, +·10 and +·10 " read " 12, 10 and 10 "
Page 507, last line, for " *africanum* " read " *africana* "
Page 508, line 1, for " (*Nematocera*) " read " (*Nematocrema*) "
Page 515, 18 lines from end for " about 20 in *greeni* " read " about 20 pairs in *greeni* "
Page 520, 8 lines from end, for " *Paleococcus* " read " *Palaeococcus* "
Page 525, line 10, for " Bull. ent. Res., **27** " read " Bull. ent. Res. **17** "
Page 619, line 21, for " Bagun " read " Gabun "

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BULLETIN
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THE TOXICITY OF DDT TO MAN AND ANIMALS.

A REPORT ON THE WORK CARRIED OUT AT THE ROYAL NAVAL SCHOOL OF TROPICAL HYGIENE, COLOMBO, AND A REVIEW OF THE WORLD LITERATURE TO JANUARY 1947.

By F. M. G. STAMMERS, M.A., M.B., Surgeon Lieutenant, R.N.V.R., and F. G. SAREL WHITFIELD, D.I.C., F.R.E.S., Lieutenant-Commander (Sp.), R.N.V.R.
Senior Entomologist to the Medical Director General of the Navy.

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Introduction.

In May 1944 the Entomological Section of the Royal Naval School of Tropical Hygiene came into existence at Colombo. Its functions were to assist in the training and organising of the Mobile Malaria and Hygiene Units, which were then being formed to take their part in the Burma and Malayan campaigns, and to provide protection against insect-borne disease and insects of importance to the general well-being of the men in the main naval bases of the East Indies Fleet.

No. 2 Base Entomological Unit was created to deal with the latter aspect and its work consisted of spraying ships and shore establishments against infestation, primarily from mosquitos, flies, bed-bugs and cockroaches and of the routine spraying of R.N. establishments in Colombo for dengue control.

The formation of this unit coincided with the provision of DDT in bulk to the Navy and the results, in terms of the additional comfort and well-being of the men, were so marked that the demands upon its services became overwhelming. In order to cope with the situation the unit was expanded and organised upon self-contained "commercial" lines, and Lieutenant (Sp.) E. D. Toner, R.N.V.R., was placed in charge. The efficient running of the unit and the excellent results obtained owe much to this officer and his assistants Sick Berth Petty Officers (S.I.) H. Rodgers and C. H. Bossley, whose enthusiasm and keenness never flagged under the most trying conditions.

The nature of the work provided excellent opportunities for the study of DDT as an insecticide under a wide range of conditions, and a central information bureau was set up at the R.N.S.T.H. in order to amass and collate the essential data.

So far as the unit in question was concerned, a job-card card index system was devised that not only enabled the officer in charge of the unit to keep pace with the work and to employ his men to the greatest advantage, but also provided a classified system of recording the essential entomological and operational data.

The work of the Entomological Section of the R.N.S.T.H. is being analysed and written up and, together with the investigations on insecticides carried out during 1945–46, will be published in due course. The entomological section of the present paper is presented on account of its importance in the interpretation of the clinical and physiological findings and no attempt is made to discuss the purely entomological aspect.

With the appointment of a Medical Officer to the school for special duties at H.Q. (F.M.G.S.), it was decided to investigate the much discussed question of the toxicity of DDT to man, a matter of some importance in view of the increasing activities of the unit. The writers in due course presented the medical findings to Professor G. R. Cameron for his comments and he immediately stressed the importance of the conditions under which the men operated. Much had been published, he pointed out, on the purely toxicological aspect, but in only very few cases had any information as to the precise degree and nature of exposure to DDT been presented, and these somewhat sketchily, upon only two or three subjects and for comparatively short periods of time.

As the investigations at Colombo had been made on a spray-operating team of 15 men, 10 of whom had been employed continuously for a period of 9 months and the other 5 for 7 months, and as their work was entirely carried out under operational conditions with a continuously high degree of exposure to DDT, it was decided to present a detailed analysis of the nature of their work and the nature and degree of their exposure to DDT, as Professor Cameron considered that this would greatly enhance the value and significance of the medical findings.

It had originally been intended to publish in a medical journal, but in view of the fact that the greatest responsibility for the safe use of DDT lies with the applied entomologist rather than with the medical man it was considered more suitable to an entomological publication.

Since the writers' return to the U.K., one of them (F.G.S.W.) has been engaged upon insecticide trials, during the course of which a method of producing a continuous-phase aerosol by electrical volatilisation was investigated. The value of this as a means of testing the toxicity of a permanent aerosol for a given period of time was taken advantage of and investigations were carried out at the Chemical Defence Experimental Station, Porton, and at the Royal Naval Hospital, Plymouth. Extracts from their findings are presented in this paper.

In accepting the rough draft of this paper for publication, Dr. W. J. Hall suggested that the time had come when a summary and analysis of the literature on the toxicity of DDT, in conjunction with the operational nature and extent of the data of the present paper, would be of value. This is accordingly presented in the hopes that it will prove of use as a work of reference and a clarification of the present somewhat nebulous position with regard to the safe use of a most important insecticide.

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The War Office for permission to quote from Interim Report No. 3, C.D.R.E., India.

The Ministry of Supply for permission to use the reports from the Chemical Defence Experimental Station, Porton, dealing with the work on the toxicity of continuous-phase aerosols carried out by them for the Navy, and in particular to Lieutenant-Colonel E. F. Edson, O.B.E., R.A.M.C., and Mr. S. H. Fryer for their collaboration and kindness in finding time and space to carry out these investigations.

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We would also like to thank Sick Berth Petty Officer W. A. Page and Sick Berth Petty Officer (L.) R. Ronaldson for their help in the examination of the men in the Colombo trials.

The Toxicity of DDT to Man under operational spraying Conditions in the Tropics.

A. Working Conditions and the Nature and Degree of the Men's Exposure to DDT.

Of the 15 men investigated, 10 had been employed continuously for a period of 9 months, and the other five for 7 months; with the exception of one Tamil, all were Sinhalese. The work of the unit consisted of the spraying of a 5 per cent. solution of DDT in grade 2 kerosene. The quality of the DDT varied but the average was approximately 75 per cent. p.p. isomer. Knapsack type sprayers were used and the solution was prepared by members of the spraying team in 40-gallon revolving drums. A mush was first formed in 4 gal. petrol tins which an operator stirred with a medium-sized stick until it was smooth and homogeneous and this was tipped into the mixing drums, to which was added the remainder of the kerosene. The men worked in pairs, one operating the sprayer for half the time while the other dealt with furniture and other loose or movable objects and vice versa. On average, a 24-hour (operating time) week was worked, much of the time being taken up with travelling, and the setting up of equipment, running repairs, etc.

Protective Clothing.

At the beginning protective clothing was issued, consisting of blue overalls, cap, tropical A.R.P. oiled silk gas cape and gloves, gum boots and gauze mouth pads. In a very short time the men had discarded the gas capes, gloves and mouth pads on account of the intense discomfort from heat, while the gum boots were frequently removed during operations, particularly in ships and on slippery surfaces as the men greatly preferred working with bare feet. All the men wore their overalls open at the neck and most of them rolled their sleeves up.

They washed their hands before the midday meal, and the majority of them washed all over at the end of the day under a cold-water tap in the open.

Degree of Exposure to DDT.

The nature of the spraying, being to a great extent indoors and much of it in confined spaces, meant that the men were frequently working in a saturated atmosphere, while many of the sprayers leaked owing to the hard treatment they received and the difficulty in obtaining spares and replacements.

The hands, forearms and upper arms and, to a lesser extent, the faces and feet, of the operators were constantly exposed to spray dripping from ceilings and splashing back from walls and to liquid solution leaking from the joints and nozzles of the sprayers. The average Ceylonese worker's idea of efficient spraying was to bring the nozzle of the sprayer as close as possible to the surface to be sprayed and to saturate the neighbourhood with competence and enthusiasm. No matter how much the undesirability of such a procedure was stressed, the team, who, in other respects became, in the course of time, expert sprayers, could never resist the temptation to saturate a surface. The results of such technique in confined spaces in the tropics may be imagined, to say nothing of the waste of insecticide. After a day's operations a white frost of DDT crystals could be seen on the exposed portions of the skin, while the overalls were frequently saturated. All attempts at making the men wear other types of protective clothing failed owing to the heat.

Table I is an analysis of the nature and environment of the work of the 15 men. in question during the period under review. The significant data have been condensed and further classified in Tables II, III and IV. The data is classified under five main groupings according to the degree of exposure to DDT, each of these is further classified according to the type of locality. The degree of exposure to DDT is indicated by the symbol ●, ranging from (5) ●●●●● to (1) ●. This has been assessed from a number of factors: spraying time, amount of insecticide used, the nature and degree of infestation and the nature of the environment. The two former factors were naturally influenced to a considerable extent by the latter two and in each case the report of the officer or petty officer in charge had a bearing upon the classification.

●●●●● indicates gross contamination, ● appreciable contamination, with varying degrees in between: even with the ● operations, hands, forearms and backs were usually soaked, and after a day's work contamination was invariably substantial, and neither of the writers can recall an occasion when the men did not return from a day's operations soaked to the skin with the 5 per cent. solution.

The Nature and Degree of Infestation.

This was one of the essential data recorded upon the job-cards and is of interest in its relation to the type of operation and the degree of exposure to DDT. It is represented by symbols thus:—

+++++ =*very heavy infestation*: with insects swarming in great numbers. Average number of insects visible or to be easily found in the immediate locality, 5,000 or more.

++++ =*heavy infestation*: with large numbers of insects visible to casual inspection. Average number visible, etc., 1,000.

+++ =*medium infestation*: with insects visible, according to species, on casual inspection, but not obtrusive; a considerable trial to the men, especially in the cases of biting species. Average number visible, etc., 200–500.

++ =*moderate infestation*: evident to inmates of the building or ship concerned but requiring some search to reveal the foci of infestation. Average number of insects visible, etc., 100-200.

+ =*light infestation*: insects evident in small numbers but frequently unsuspected by inmates of quarters, potentially dangerous. Average number of insects visible, etc., 5-100.

Meteorological Data.

The mean average shade temperature during the period of investigation was 80°F.

The relative humidity recorded from Naval sources was 79 per cent. The records of the Colombo Observatory give the following figures :—

Average shade temperature for 11 years ... ... 80·4°F.

Average rainfall for 31 years ... ... 90·8 inches.

(May 15 ins., October 13·2 ins., November 12·3 ins. The remaining months averaging 5·5 ins.)

Average relative humidity for 20 years :—

(A) 74 per cent.—Max. 78 per cent., June, July.
Min. 68 per cent., February, March.

(B) 90 per cent.—Max. 93 per cent., March, November.
Min. 87 per cent., June, July, August.

(A)=Monthly means obtained from dry and wet-bulb readings at 9.30 a.m. and 3.30 p.m.

(B)=Monthly means obtained from the minimum dry and wet-bulb readings. The minimum dry and wet bulb temperatures do not necessarily occur at the same instant and a consequent uncertainty is introduced. The evidence of thermographs shows that this is very small and as the relative humidity at night differs considerably from that during the day, the (A) figure alone gives a very incomplete statement of conditions as a whole.

The temperature and humidity in many of the ships and buildings sprayed were appreciably higher than those generally prevailing at the time and conditions were, at times, exceedingly trying.

The term " barrier spraying " used in the table denotes the treatment of an area, usually surrounding the locality to be controlled, in which all types of terrain, plantation, scrub, jungle, undergrowth, grass, open ground, etc., were thoroughly sprayed as a protection against adult mosquitos in particular and insects in general. This method of control was evolved, in consultation with Col. J. W. Scharfe, R.A.M.C., of the Medical Advisory Division, S.E.A.C., as the best standardised method of protection for barracks, camps, etc., situated in prolific breeding country in which general control measures were impossible. On the whole very satisfactory results were obtained, and under operational war conditions it was by far the easiest and most effective method of large-scale protection for man-power in the mass.

A Banda is a type of native hut, rectangular in shape, of varying length, built of palm thatch and bamboo. Many were erected with brick or concrete walls and thatched roofs, or half-brick half-thatch walls to house Naval personnel, their capacity varying from 10 to 50 or more personnel.

The Insects referred to were :

Mosquitos : a majority of *Culex fatigans*, Wied., and *C. gelidus*, Theo.
a sprinkling of *Aëdes albopictus*, Skuse.
occasional pockets of *Aëdes aegypti*, L.

Flies : *Musca sorbens*, Wied., with occasional *Lucilia caesar*, L., and *Calliphora.*

Cockroaches : mostly *Blatta orientalis*, L., with occasional *Periplaneta americana*, L., and a very few *Blattella germanica*, L.

TABLE I.

Basic data on control measures carried out by the No. 2 base Entomological Unit, R.N.S.T.H., Colombo, during the period of the clinical and physiological investigations, with particular reference to the degree of exposure to DDT undergone by the men.

In all cases, except where otherwise stated, the formula was a 5 per cent. solution of commercial DDT in grade 2 kerosene.

Equipment used, except where otherwise stated, was the "Ross"-type "Four-Oaks" knapsack sprayer with the No. 2 spray nozzle.

The commonest insects abbreviated thus:

Mosquitos ... ... M.
Flies ... ... F.
Bed-bugs ... ... B.
Cockroaches ... ... C.

Type of Locality	Nature and Degree of Infestation	Nature of Operations	Gallons of Insecticide	No. of Operators	Spraying Time in Hours
Degree of Exposure to DDT ● ● ● ● ●					
8 M.L.s	C. +++	Entire ships sprayed	24	10	2·5
Submarine depot ship	C. ++++ B. ++++ Ants +++	Converted merchant ship. Holds, cabins, store-rooms sprayed	125	8	18
Ditto	C. +++ F. ++ Ants ++	All parts ship infested. Sprayed throughout where feasible	104	12	6
*Merchant Vessel ...	B. +++ C. +++	Officers' and crews' quarters, passenger accommodation, walls and furniture	56	12	4·5
* At request of Sea Transport Officer.					
Degree of Exposure to DTT ● ● ● ●					
Cruiser	B. +++ C. +++	Mess decks, offices, sick bay, officers' quarters, wardroom, stores, walls and furniture sprayed	40	12	2·5
Destroyer	C. ++	Mess decks, galley, wardroom and all living quarters	48	12	3
Ditto	B. ++++ C. ++++	Living quarters, mess decks, wardroom walls and furniture	32	8	3
Submarine	C. ++	All parts of ship	12	5	3
Ditto	B. +++ C. +++	All parts of ship	6	4	1
Frigate	C. +++	Living quarters, offices, stores, wardroom, galley	40	8	2
Ditto	C. ++++	Mess decks, wardroom, officers' cabins, offices, stores	30	6	2·5
Ditto	C. nymphs breeding up after HCN fumigation +++	12 messes, officers' quarters, cold storage and food stores	40	10	3

TABLE I—*continued.*

Type of Locality	Nature and Degree of Infestation	Nature of Operations	Gallons of Insecticide	No. of Operators	Spraying Time in Hours
Degree of Exposure to DDT ●●●●—*continued.*					
Last re-sprayed three months later	C. ++++	All living quarters, galley, storerooms, flats	44	8	2
Corvette	C. +++	Mess decks, officers' cabins, storerooms, galleys	64	8	2
Sloop	C. +++ B. ++	Mess decks, cabins, ward room, galleys, stores, furniture, bedding, etc.	35	10	1·5
Minelayer	C. +++ B. +	Living quarters, galley, wheel house, store rooms	14	4	1·5
8 M.L.s	C. ++ B. +	Living quarters, wheel house, bunks, bedding	36	12	1·75
5 M.L.s	C. +++	Living quarters, wheel houses	20	6	2
3 M.L.s	C. +++	Living quarters, wheel houses	14	6	1·5
Depot Ship	C. ++	Mess decks, cabins, galleys, workshops, stores	74	14	4
Ditto	C. +++ B. +++	Holds, storerooms, native crews' quarters.	45	12	4·5
Cable Ship	C. +++ B. +++	Mess decks, cabins, stores, cable tanks, bulkheads, deckheads, bedding, bunks	35	6	2
Merchant Ships ...	C. ++++	Mess decks, flats, cabins, galleys, stores	76	16	2·5
R.N. Boat Yard ...	M. +++	Cabins and wheel houses of all boats in yard	25	12	1·75
Large base hospital...	F. ++++ B. ++++	Living quarters, dining halls, galleys, wards, officers' block, barrier spraying throughout area	200	12	9
Above re-sprayed after 3 months	F. ++ C. +++	Hospital sprayed throughout, all furniture and bedding; residual and barrier	304	14	9
W.R.N.S. quarters ...	B. +++	House walls, beds, bedding, furniture, bandas	105	12	5·5
Ditto	M. ++++	Breeding in neighbourhood, residual on walls, barrier	80	12	5·5
European barracks ...	B. ++++ F. ++++	Residual on walls, furniture, bedding	295	13	21
Native barracks ...	B. ++++	Galleys, offices, quarters, furniture, bedding	70	20	2

TABLE I—*continued.*

Type of Locality	Nature and Degree of Infestation	Nature of Operations	Gallons of Insecticide	No. of Operators	Spraying time in Hours
Degree of Exposure to DDT ● ● ● ●—*continued.*					
Subsidiary European barracks	F. ++++	All barrack area, bandas, galleys, barrier	120	16	4
Ditto	B. ++++ F. ++++	Brick bandas, galley, barrier	88	12	6
Miscellaneous* ...	B. +++ F. +++	Residual in galleys, dining halls, bandas, barrier round galley	180	10	16
Ditto	B. +++ F. ++++	Walls, furniture, bedding, nets, barrier round bandas, offices and galley	308	12	16
Ditto	B. ++ F. +++	Office furniture, workshops, canteen, galley	60	8	8
Ditto	B. +++ F. ++++	Bedding store, P.O.'s mess, officers' mess, beds, residual in mess hall	107	8	14
Camp†	F. ++++	Residual in galleys, dining halls, bandas, external breeding areas and barrier	235 & 145 DDT/malariol	12	24
Service men's club ...	B. +++++	Furniture, beds, walls	95	12	4
Victualling and clothing store	Termites ++++	Crates and boxes stripped and sprayed, clothing sprayed	44	2	30
Degree of Exposure to DDT ● ● ●					
Destroyer	C. +++	Galley, wardroom, living quarters	14	8	2·5
Frigate	C. ++	Mess decks, flats, stores, galleys	25	10	1·5
Corvette	C. ++	Mess decks, wardroom, quarters ...	16	8	1·5
Minelayer	B. ++ C. +++	Mess decks, galleys, officers' quarters, wheel house, washplaces, storerooms, hold	14	4	1
Trawler	B. + C. ++	Mess decks, wardroom, cabins, galley, stores	6	8	0·5
Salvage Ship ...	M. *Aëdes aegypti* breeding in ship. Adults ++++	All breeding places, laundry residual for adults	2	2	1
Pilot Parent Ship ...	F. ++++	In holds from ballast, holds sprayed	10	8	2
Two Motor Fishing Vessels	B. ++++	Living quarters and new tongued and grooved bulkheads	6	4	1

* Wireless stations, etc.

† Very bad hygiene, situated in middle of coconut plantation.

TABLE I—*continued.*

Type of Locality	Nature and Degree of Infestation	Nature of Operations	Gallons of Insecticide	No. of Operators	Spraying time in Hours
Degree of Exposure to DDT ● ● ●—*continued.*					
4 M.L.s	C. +++ B. +	All living quarters in each ship	16	8	1
3 M.L.s	C. +++ B. +++	Wheel houses, living quarters	12	6	1
1 M.L.	C. ++	Quarters and stores	6	4	1
Ditto	B. ++	Quarters and stores	6	2	1·5
Ditto	C. ++++	Quarters and galleys	8	4	0·75
Miscellaneous* ...	B. +++ C. ++	Living quarters, messdecks, storerooms	14	6	1
Ditto	B. +++ C. +++	Living quarters, messdecks, galleys, storerooms	20	8	2
Ditto	B. + C. +++	Mess decks, galleys, cabins, stores, wardroom	15	8	1
Officers' block in large base hospital	M. ++ F. ++	Residual on walls of wards and galley, barrier	32	16	2
"A" block in large hospital	F. +++ B. ++	All wards, walls, beds, lockers, chairs, barrier	72	8	4·5
V.A.D.'s quarters ...	M. ++ B. ++	Two houses and three bandas residual on walls, furniture and barrier	110	12	5
Sisters' quarters ...	B. +++ M. complaints	Walls, beds, chairs, screens	84	8	4·5
R.N. ambulance train	B. many complaints	8 coaches treated, settees, sleepers, etc.	12	6	0·75
Galley and dining halls	F. +++ B. ++	Galleys, serveries, stores, halls, all furniture	30	8	1·5
Ditto	F. ++	Carried out by request as additional precaution in typhoid outbreak, residual	42	6	3
Galleys	F. +++ B. ++	Walls and furniture, barrier	50	6	2·5
Canteen and galley ...	B. ++++ F. ++	Kitchens and all furniture, residual	35	12	2·5
Canteen	B. +++	Furniture, walls, church pews, school desks	36	8	1·75
W.R.N.S. mess ...	M. +++	House and bandas, offices, stores and beds, barrier	196	22	5·5
Admiralty civilians' mess	M. +++ B. +++	Walls, furniture, mosquito nets, barrier	135	8	6
W.R.N.S. mess ...	B. ++ M. ++ Ants +++	House and bandas residual, beds	124	12	5·5
Mess	M. ++	Walls and furniture, barrier	116	16	3

* Miscellaneous ships, local Register.

TABLE I—*continued.*

Type of Locality Case No.	Nature and Degree of Infestation	Nature of Operations	Gallons of Insecticide	No. of Operators	Spraying Time in Hours
Degree of Exposure to DDT ● ● ●—*continued.*					
Group of messes ...	F. +++ B. + M. complaints	Walls, furniture, beds, stores, galley, dining halls, barrier	116	12	4
Admiralty civilians...	Pre-occupation spraying	Internal walls, furniture, bedding, outer walls	96	12	3
W.R.N.S. mess ...	M. } complaints F. }	4 houses, bungalow, walls, furniture, barrier	74	14	3
Ditto	M. } complaints F. }	Furniture, internal walls, barrier	80	14	3
Ditto	B. ++	2 houses, 5 bandas, internal residual	56	6	3·5
Last mess, 3 months later	C. +++ M. +	General internal, barrier	56	8	4
W.R.N.S. mess ...	B. +++	House, 5 bandas, all furniture, walls	50	6	4
Civilian mess ...	F. ++ B. ++ M. ++	Galley, quarters, walls, furniture, barrier	45	8	2
Fire and police quarters	M. ++ B. ++	Quarters, offices, galleys, dining rooms, furniture, static water tanks	42	6	1·75
W.R.N.S. mess ...	F. +++ M. +++	Internal walls, barrier	40	6	3
Living quarters, Miscellaneous	B. ++++ F. +++	All furniture, galley, chairs in cinema	120	12	3·5
Ditto	B. +++ F. ++ C. ++ M. +	Bandas, galley, dining halls, W/T station, furniture	90	8	4
Ditto	B. ++	Bedding and furniture	70	12	3
Ditto	B. +++	All office furniture	58	6	8
Camp*	M. +++ F. ++++ B. ++	Internal residual and barrier throughout camp	812	Varied from 12–16	24
Ditto	F. +++ M. ++ *Dinoderus* sp. +++	Bandas, furniture, galleys, messes, barrier in places	160	16	4·5
Ditto	B. +++ F. ++ M. ++	Bandas, tents, buildings, furniture, barrier in parts	124	20	2·75
Ditto	F. ++++ M. +++	Bandas, tents, huts, internal and barrier	142	12	4
Ditto	B. ++++ F. ++	Bandas, internal and all furniture	100	12	4
Ditto	B. ++++	Furniture and beds	85	10	6
Ditto	Chironomids ++++	Bandas, galley, wardroom, dining halls, barrier	58	8	2·75
Ditto	B. +++++	Beds, walls, nets	40	4	3
Club	B. ++++	Beading on walls, all furniture and beds	65	10	6·5

* Special case, very bad hygiene conditions.

TABLE I—*continued.*

Type of Locality	Nature and Degree of Infestation	Nature of Operations	Gallons of Insecticide	No. of Operators	Spraying time in Hours
Degree of Exposure to DDT ● ● ●—*continued.*					
Institute	B. +++	Galleys, bandas, furniture	52	16	2
Ditto	B. +++ C. ++	Furniture, kitchen cupboards	20	4	2
Office	B. +++	New walls, tongue and groove, all furniture	40	8	3·5
Boatyard	Chironomids +++++ M. +	All buildings internal and external residual, barrier along lakeside	60	8	3
Clothing store ...	*Dinoderus* sp. ...	Arresting attacks on racks and shelves, protective treatment of remainder	120 of DDT/kerosene/varnish solution	12	4
Ditto	F. +++	Walls and shelves	12	4	1
Large Hotel	B. ++++	All furniture, bedding	86	12	5
Medium Hotel ...	F. ++++ B. ++	Kitchen walls, furniture, beds, chairs in lounge	76	12	3·5
Ditto	B. ++++	All beds and furniture	68	14*	3
Large Hotel	B. +++	Chairs and tables in public rooms	50	12	6
Ditto	F. +++	Kitchens, refuse dumps, servants' kitchen	20	6	2
Degree of Exposure to DDT ● ●					
2 Hospital wards ...	F. ++ M. ++	Walls, beds, lockers	24	8	1
1 ward	M. ++	Walls and some furniture	20	8	2
2 wards	M. complaints F. +	Walls, beds, furniture	14	8	1
Sick bay	M. ++	Walls and verandah	16	4	1
Sick quarters ...	Chironomids ++ B. +	Walls and two ceilings, some beds	10	6†	1
Galley	F. ++	Walls and beef screens	9	4	1·5
Canteen	M. +++	Walls, some furniture	20	6	1
Ditto	B. ++	Chairs, tables, camp beds	16	6	1
Ditto	B. ++++	Chairs, tables	15	6	1·5
Ditto	B. +++	Chairs, tables	12	8	1
Ditto	B. +++	Chairs, tables, lower walls	12	4	1
Ditto	B. +++ C. +++	Empty rooms, stores and kitchen	8	4	1·5
W.R.N.S. quarters ...	F. ++	Houses, bandas, galleys, walls, internal and external	64	16	2

* Six knapsack sprayers and one motor-driven air compressor sprayer.
† Three knapsack and one single-acting "Eclipse" pump sprayer.

TABLE I—*continued.*

Type of Locality	Nature and Degree of Infestation	Nature of Operations	Gallons of Insecticide	No. of Operators	Spraying time in Hours
Degree of Exposure to DDT ● ●—*continued.*					
W.R.N.S. quarters ...	M. ++ F. +++	Houses, bandas, barrier	50	6	2
Ditto	M. ++	Walls, dining hall, galley, stores, internal and external	48	8	2
Admiralty civilians...	M. ++	All walls internal, barrier	36	8	2
W.R.N.S. quarters ...	Pre-occupational spraying	Residual throughout house, bandas and galleys	34	6	2
Ditto	B. +++ Ants. +++	Bungalow and several bandas, residual on lower walls and furniture	26	8	1·75
Ditto	F. +++ B. ++	Dining hall, galley, mail office, regulating office	20	6	1·5
Ditto	F. +++	Galley, servery, walls, tables, partial barrier	16	4	1
Ditto	B. +++	Furniture and lower walls	16	8	2·25
Ditto	F. +++ B. ++	Galley, chairs, barrier	13	4	1
Barracks	B. +++	Bedding and all furniture	30	6	2·5
Ditto	B. ++++	Bedding and all furniture	15	6	1·75
Camp	M. ++ F. +++ Ants. +++	Huts, latrines, stores, barrier throughout	160	22	3·5
Ditto	B. +++ F. +++ M. ++ Ants. ++	All bandas, galleys, offices, barrier	80	16	2·25
Ditto	F. +	Latrines, galley, local barrier	26	4	2·5
Ditto	F. +++	Farm buildings, internal	16	4	2
Institute ...	M. +++	Internal walls and barrier	32	16	1·5
Club	B. ++	Furniture and walls partly	20	12	1·5
Institute	B. ++	Lounge furniture, galley	16	6	1
Store	M. complaints B. ++ C. +++	Walls and fittings, furniture	16	6	1
Store	M. ++	Walls, internal and external	70	12	3
Offices	B. ++	Chairs and desks	30	8	1·5
Film centre	M. ++ B. ++	Office furniture, cinema and work room	24	6	1·5
Mail office	M. ++ B. +	Furniture, counters, walls, sorting racks	24	12	1
Office	B. +++	Chairs and desks	20	4	2
Ditto	B. +++ C. ++	Furniture and walls	20	6	1·5

TABLE I—*continued.*

Type of Locality	Nature and Degree of Infestation	Nature of Operations	Gallons of Insecticide	No. of Operators	Spraying time in Hours
Degree of Exposure to DDT ● ●—*continued.*					
Offices	M. +++	Walls residual, gutters, drains	20	4	2·25
Planning rooms ...	B. ++	Furniture and lower walls	17	4	2
Office	B. +++	Chairs, tables, desks	16	12	1·5
Ditto	B. ++++	All furniture, camp beds	15	8	2
Office, drawing office	M. +++	Lower walls, under chart tables and drawing tables	12	4	1·5
Office	B. +++	Chairs and desks	12	4	1·75
Ditto	B. +++	All furniture	8	4	1
Small commercial hotel	B. +++	All beds treated and surrounds	37	8	2
Ditto	B. ++	Furniture and beds	25	8	2
Cinema	B. +++	All seats and chairs	50	12	2
Ditto	B. ++++	All seats and chairs	47	8	2·5
Ditto	B. ++++	Seats, surrounds and wooden flooring	45	12	1·75
Ditto	B. ++	Seats and surrounds	40	12	2
Ditto	B. +++++	All seats	40	6	3·75
Ditto	B. +	All seats	35	12	1·25
Ditto	B. +++	All seats	32	12	1
Degree of Exposure to DDT ●					
Up-country sick-bay	B. complaints	General residual	7	2	0·5
R.M. Mess	B. +++	Bedding, chairs, nets	5	2	0·5
Officers' mess ...	C. ++++	Focus of infestation, galley	2	1	0·33
W.R.N.S. quarters in W/T station	B. +	Beds only	10	2	0·75
Camp	F. ++++	Galley and area only	8	4	0·5
Ditto	F. +	Wardroom, galley walls	9	4	0·75
Institute	B. +++	Chairs and tables	8	4	0·5
Office	M. complaints	Walls and furniture	8	4	0·75
Ditto	B. complaints	Walls and furniture	8	4	0·5
Photographic office...	F. } complaints M. }	Walls and furniture	8	4	0·5
Office	B. ++	Chairs and desks	6	8	0·5
Chart room	M . complaints	Walls where no charts	12	8	0·75
W/T room	B. ++ M. complaints	Furniture and some walls	4	2	0·5
Office	M. ++ F. ++ B. +	Furniture and walls	10	4	0·75
Censor office ...	B. +++	Desks and chairs	5	6	0·33
Office	B. +++	Furniture	15	8	0·74
Ditto	B. +++	Furniture	9	4	0·75
Ditto	B. } complaints M. }	Furniture and walls	8	8	0·5

The drop in quantity of insecticide used under ships (Table I) is accounted for by the close and difficult conditions under which the operations were carried out, which also accounted for the high degree of exposure to the insecticide.

Table II shows the relation between the type of locality, the degree of exposure to DDT and infestation. It will be noted that ships and living quarters provide the highest degree of contamination by DDT owing to the confined working conditions. The exceptions to be found under other locality headings are due to the combination of long spraying time and the large amounts of insecticide used, as shown in Table III.

TABLE II.

Relation between type of locality, degree of exposure to DDT and infestation.

Names of insects abbreviated thus :—

Flies	F.	Ants	A.
Cockroaches	C.	*Dinoderus* sp.	D.
Bed-bugs	B.	Chironomids	Ch.
Mosquitos	M.	Termites	T.

Type of Locality	No. of Cases	Degree of Exposure to DDT	No of Cases	Nature and Degree of Infestation						
				Insect	Total No.	5 +	4 +	3 +	2 +	1 +
Ships	40	●●●●●	4	F. C. B.	1 4 2		 1 1	 3 1	1 	
		●●●●	20	M. C. B.	1 19 9		 4 1	1 11 5	 4 1	 2
		●●●	16	F. M. C. B.	1 1 12 9		1 1 1 1	 6 3	 5 2	 3
Hospitals ...	13	●●●●	2	F. C. B.	2 1 1		1 1	 1 	1 	
		●●●	5	F. M. Ch. B.	2 3 1 4			1 1	1 2 1 2	 1 1
		●●	5	F. M. B.	2 3 1				1 3 	1 1
		●	1	B.	1					1
Canteens and Galleys ...	12	●●●	5	F. B.	4 4		 1	2 1	2 2	
		●●	7	F. M. C. B.	1 1 1 5			 1 1 4	1 1	
Living Quarters (Messes, Barracks and Miscellaneous)	43	●●●●	10	F. M. B.	7 1 8		5 1 3	2 4	 1	
		●●●	18	F. M. C. B.	3 8 1 7			2 3 1 2	1 4 4	 1 1

TABLE II.—*continued.*

Type of Locality	No. of Cases	Degree of Exposure to DDT	No. of Cases	Nature and Degree of Infestation						
				Insect	Total No.	5 +	4 +	3 +	2 +	1 +
Living Quarters (Messes, Barracks and Miscellaneous) —*cont.*	43	●●	12	F.	5			4	1	
				M.	3				3	
				A.	1			1		
				B.	6		1	3	2	
		●	3	C.	1		1			
				B.	2			1		1
Camps	15	●●●●	1	F.	1		1			
		●●●	8	F.	5		2	1	2	
				M.	4			2	2	
				Ch.	1		1			
				D.	1			1		
				B.	5	1	2	1	1	
		●●	4	F.	4			3		1
				M.	2				2	
				A.	2			1	1	
				B.	1			1		
		●	2	F.	2		1			1
Clubs and Institutes ...	8	●●●●	1	B.	1	1				
		●●●	3	C.	1				1	
				B.	3		1	2		
		●●	3	M.	1			1		
				B.	2				2	
		●	1	B.	1			1		
Stores	5	●●●●	1	T.	1		1			
		●●●	2	F.	1			1		
				D.	1			1		
		●●	2	M.	2				1	1
				C.	1			1		
				B.	1				1	
Offices ...	24	●●●	1	B.	1			1		
		●●	12	M.	4			2	2	
				C.	1				1	
				B.	10		1	5	3	1
		●	11	F.	2				1	1
				M.	6				1	5
				B.	7			3	2	2
Workshops ...	1	●●●	1	M.	1					1
				Ch.	1	1				
Hotels ...	7	●●●	5	F.	2		1	1		
				B.	4		2	1	1	
		●●	2	B.	2			1	1	
Cinemas ...	7	●●	7	B.	7	1	2	2	1	1
Totals ...	175					4	40	98	72	27

Totals for insects: Bed-bugs—104, Flies—45, Cockroaches—42, Mosquitos—41, Chironomids—3, Ants—3, *Dinoderus*—2, Termites—1.

As has been pointed out ships and living quarters provide the bulk of cases of heavy contamination from DDT, the few exceptions are shown in Table III.

TABLE III.

Degree of Exposure to DDT ●●●●

Type of Locality	Case Number	Gallons of Insecticide	Spraying Time	Infestation
Hospital	1	200	9 hours	F.++++ B.++++
Ditto	2	304	9 ,,	F.++ C.+++
Camp	1	380	24 ,,	F.++++
Club	1	95	4 ,,	B.+++++
Store	1	44	30 ,,	Termites++++

The first three cases are localities where large areas, combined with barrier spraying, involved considerable time and material. The club really qualifies, from the point of view of spraying conditions, as a living quarter, while the store, a victualling and clothing unit, was badly infested with termites and although crates, boxes, bales of clothing and cloth had all to be moved, stripped and sprayed, only two men could be assigned to the work with a consequent lengthening of the spraying time.

TABLE IV.

Summary of Operational Data in relation to the Degree of Exposure to DDT.

Degree of Exposure to DDT	Number of Cases	Average No. Operators	Average amount of Insecticide used	Average Spraying Time
●●●●●	4	10·5	77·2 gallons	7 hours 42 minutes
●●●●	35	9·2	90·4 ,,	6 ,, 24 ,,
●●●	64	9·0	68·2 ,,	3 ,, 12 ,,
●●	54	7·8	28·6 ,,	1 ,, 42 ,,
●	18	4·5	7·8 ,,	35 ,,

To elaborate a little further, during the 9 months under review 175 spraying operations were carried out, involving the expenditure of 9,402 gals. of DDT/kerosene solution with a total spraying time of 566 hours. Averaging both the 9-month and the 7-month groups, the figures per spraying operator are 8·5 men working for 3¼ hours and using 52·5 gals. of insecticide.

Each man had, therefore, theoretically assisted in 175 operations with an average spraying time of 3¼ hours and a consumption of 6·15 gals. of insecticide and at the end of the 9-month period each man had completed 566 hours spraying and expended 626·8 gals. of insecticide. The total output of the unit was 7,490 man hours of control operations.

B. The Medical Investigation.

Of the 15 men who were investigated, 10 had been employed for a period of 9 months and the other 5 for 7 months. Each man was questioned as to his general fitness and received a full clinical examination, including blood pressure estimations. Particular attention was directed to the skin, mucous membranes and the nervous system; urine was examined for albumen and liver function and haemoglobin estimated, the former by the oral Hippuric acid synthesis test, the latter by the method of Sahli.

The liver function test and haemoglobin estimations were also carried out on a number of Sinhalese of approximately the same age and from the same districts, all of whom were employed at the R.N.S.T.H. on duties other than those of handling DDT and kerosene.

Two months later red and white cell counts, including differential white cell counts and examination of stools for parasitic ova, worms and cysts were made on 12 of the same men, 9 of whom had been engaged on spraying for 11 months and the remaining 3 for 9 months.

All of the men were Sinhalese with the exception of one Tamil. Their ages ranged from 21 to 40 years, averaging 26·4 years. With the exception of one man who complained of an increased frequency of micturition with some pain during the act, each said that he felt perfectly well. The overseer in charge endorsed their statements and maintained that their work had not deteriorated in any way during the period of their employment.

Some of them were in the habit of smoking a mixture of hemp and tobacco and although an exact estimate could not be made it seemed that only an infrequent use was made of the former drug.

Their physique seemed a little better than that of other young men in Colombo and with the exception of one *Tinea cruris* their skins appeared healthy. Some of them had experienced skin irritation with burning sensations which went into vesicle formation and patchy desquamation when they first started spraying. The condition cleared up within a few days and was not severe enough to stop them working. Skin tests with grade 2 kerosene gave a similar picture that was typical of kerosene dermatitis seen in European workers doing similar operations with pyrethrum-based sprays. There was no repetition of these initial reactions to kerosene except on one or two occasions when, either due to carelessness or equipment faults, contamination had been exceptionally severe. Mucous membranes appeared normal but two men showed a mild degree of conjunctival congestion. No abnormal physical signs were found on examining their chests.

Blood pressure readings were as follows :—

Systolic ranging between 90–138 mms. (below 100 in one man). Average 111 mms.

Diastolic ranging between 61–82 mms. Average 70 mms.

With the exception of one slightly enlarged liver, nothing abnormal was found on palpating the abdomen. There was no evidence of tremor nor were there any other findings indicating a departure from the normal.

Liver Function Test.

The method used for ascertaining the excretion of hippuric acid was that of Quick (1933). During the four-hour period that succeeded the 6 gm. dose of sodium benzoate 14 of the men excreted 2·4 gms. or more of hippuric acid (calculated as benzoic acid), the average value for the latter being 3·7 gms. The remaining man excreted 1·4 gms. In view of this low figure the test was repeated on him one week later with a similar result. Six weeks later, however, a third test gave a figure that was well above the average for the rest of the men. Normal results were obtained with 15 controls whose average excretion of hippuric acid was 3·1 gms.

Haemoglobin Estimation.

This was made by the simple calorimetric method of Sahli (100 per cent. being equivalent to 14 gms. of haemoglobin per 100 ccs. of blood). The average haemoglobin percentage of 74 was almost identical with that of a control series of 22 men and varied between 63 per cent. and 86 per cent.

Blood Counts.

The following cell counts were obtained :—

Red cells : from 3·3–5·3 millions per cub. mm.

Total white cell counts varied between 5,200–15,200 per cub. mm. and averaged 8,400. No abnormal cells were seen.

The differential white cell counts gave the following figures (Table V). To this table are added the red cell figures.

TABLE V.

Subject	Total red blood cells	Total white blood cells	Polymorphonuclear leucocytes	Lymphocytes	Eosinophiles	Monocytes	Basophile
1	4,090,000	15,200	10,488	4,028	532	152	0
2	4,530,000	7,000	2,450	2,730	1,540	210	70
3	5,060,000	8,400	4,284	3,234	840	42	0
4	3,600,000	10,400	5,200	4,420	676	104	0
5	4,280,000	9,400	3,760	3,478	2,115	0	47
6	4,380,000	7,800	2,547	3,822	1,131	234	39
7	4,430,000	5,400	2,754	2,079	540	27	0
8	3,310,000	9,800	5,880	2,450	1,274	98	98
9	4,190,000	5,200	2,808	1,820	416	156	0
10	4,300,000	7,000	3,290	3,220	420	70	0
11	3,420,000	7,000	3,220	3,045	560	175	0
12	5,290,000	8,600	3,440	4,773	258	129	0

Urines.

With one exception the urines were free from albumen. The urine that contained the albumen also contained pus cells in moderate numbers.

Stools.

Every stool was examined using the simple salt flotation method with the following results :—12 stools contained *Ancylostoma* ova ; 6 stools contained *Trichuris* ova ; 4 stools contained *Ascaris* ova.

Discussion.

The congestion of the conjunctivas that was observed in two men may have been caused by DDT and kerosene ; this sign is, however, a common finding in those who are addicted to hemp. Chopra & Chopra (1939) found that it was present in 72 per cent. of over 1,200 addicts examined.

The finding of a slightly enlarged liver is in no way remarkable in view of the many causes of such a condition in the tropics.

The hippuric acid synthesis test was abnormal in one man on two occasions. No ready explanation can be given for this finding but, because of the return to a normal figure six weeks later, too much significance should not be attached to it. Other factors were involved, for the man, an opium addict, stated that during this time he was endeavouring to substitute hemp in its place. He gave not only a history of several months increased frequency of micturition, but his urine contained albumen and pus cells, while his total white cell count was 15,200.

The differential white cell counts are in conformity with those of healthy individuals recorded in recent years at the General Hospital, Colombo (personal communication from Dr. G. S. W. De Saram). They show a low normal polymorphonuclear count which might possibly be due to a deficiency of one or more dietary factors such as iron (Forbes, Johnson & Consolazio, 1941), or vitamin A (Abbot, Ahmann & Overstreet, 1939). It is felt that none of the above findings can be ascribed to the action of DDT.

Conclusions.

Fifteen men attached to the R.N.S.T.H., Colombo, were investigated to ascertain whether they had been harmed by daily contact with a 5 per cent. solution of DDT in kerosene. They had been exposed for many months to heavy and continuous contamination, considerably greater than any so far published. The clinical and special investigations and their general demeanour and labour output indicate that they had received no ill effects. They were keen on their work, absenteeism seldom occurred, and a considerable *esprit de corps* existed in the team.

The Toxicity of a continuous-phase Aerosol of DDT to Man and Animals.

During the course of insecticide trials recently carried out for the Navy, a method of producing a continuous-phase aerosol of DDT by means of electrical volatilisation was investigated.

In view of the fact that all previously known aerosols remain in the air for relatively short periods of time, it was decided to take the opportunity to study the behaviour of a continuous aerosol. Trials were accordingly arranged at the R.N. Hospital, Plymouth, and six units were sent to the Chemical Defence Experimental Station, Porton, who kindly consented to investigate with their greater facilities. The work at Porton was done on a laboratory scale while that at Plymouth was conducted under normal working conditions.

A full description of the method and the entomological results will be published in due course ; it will suffice here to state that the instrument consists essentially of a circular container, housing a thermostatically controlled electrical heating element which surrounds a metal cup containing DDT. When the current is switched on the DDT is volatilised and disperses into the surrounding atmosphere.

Final data from the physical and chemical investigation are not yet to hand but it can be stated that the average output of one unit is in the region of 400 mg./24 hours with a median particle size of approximately $0{\cdot}5\,\mu$ at the mouth of the cup and an average of between $0{\cdot}5$–$5{\cdot}0\,\mu$ at normal dispersal points in a room of some 5,000 cu. ft. The instrument can be left on continuously and many units have been operating successfully for 2–3 months.

TOXICITY TO MAN.

A. Porton (Extracts from Reports submitted by C.D.E.S., Porton).

Test Room : brick and plaster rectangular-sectioned room of 4,000 cu. ft., one door in use, a further door and fireplace sealed off to restrict incidental ventilation, windows sited at either side of the room, floor partly carpeted concrete.

Siting of Units : 3 volatilising units were fixed on the walls at a height of 7 ft. above the floor and 2 ft. below the ceiling, about 8 inches away from the wall.

Description of test: The units were charged with technical grade DDT (80 per cent. p.p. isomer) and switched on from the 9th August until 17th August. A 4-day build-up period was allowed before the men entered the room for the 5-day exposure period. The actual test was carried out by permitting six Service (R.N.) volunteer subjects to occupy the room from 9.30 a.m. to 12.30 and from 2.30 to 4.30 p.m. for 5 consecutive days, that is a total of 27½ hours occupation in 5 days of a 4,000 cu. ft. room with three DDT volatilising units in continuous operation.

Subjects of the test: These were medically fit men; they were examined clinically before the test and at intervals before, during and for 4 days after the test their blood counts were determined and urine analysis carried out.

In order to prevent any possibility of the development of psychogenic complaints full amenities were provided, games, reading, radio, etc., and in order to prevent their developing suggestible complaints the clinical examination was not repeated, being withheld until there was any type of single or generalised complaint. The subjects, in fact, lived normally in the room, along with a supervisory experimental assistant, the only abnormal factor introduced being the routine blood and urine tests.

Ventilation: This was clearly of paramount importance to both the results of the test and the comfort of the inmates of the room. The intention was to provide restricted ventilation such that conditions in the room were humanly acceptable and yet would not dissipate the DDT vapour unduly.

During the pre-test 4 days the room was completely closed, without direct ventilation. During the 5-day occupation period the room was kept closed from 9.30 until 10.30 after which from 10.30 until 4.30 ventilation was permitted by opening two small fanlight windows, one at either side of the room, hinged at the top, and closed again at 4.30 until the next morning. The actual ventilation was recorded by means of a small cup-anemometer. The windows were of 1·32 sq. ft. open area and standardised readings, in two positions at each window, were from nil up to 2·1 ft./sec. Converted into cu. ft./minute volume, the ventilation occurring during 12 determinations was as follows :—

55, 101, 11·5, 12, 83, 0, 0, 162, 69, 4·6, and 4·6 cu./ft. minute.

This very restricted ventilation resulted in a slight sensory evidence of occupation being present in the day time, not sufficient to cause complaint but undoubtedly serving to prevent excessive dissipation of atmospheric DDT.

Temperature and humidity figures for the period August 13–17 were as follows :—

D.B.T. Max. 65–71°F. R.H. 65–74 per cent.
,, ,, 57–66°F. ,, 74–88 ,,

Results of Test.

The results were entirely negative. There were no complaints of any kind from any of the subjects either during the exposure period or during the four post-test days when they were detained for final observation. No effects were complained of or were determined in the eyes, nose, throat, skin, senses or general condition. The results of the serial examinations on each subject are given in Table VI (abbreviated here).

TABLE VI.

Subject	R.B.C.	W.B.C.	Hgb. Per cent.	Polys. Per cent.	Lymphs. Per cent.	Monos. Per cent.	Eosins. Per cent.	Basos. Per cent.	Date.
1	4,550,000	7,000	94	57	31	9	2	1	12th
	4,200,000	7,800	85	50	32	6	3	1	17th
	4,800,000	6,400	96	62	34	3	1	1	21st
2	4,600,000	9,200	90	60	25	12	3	0	12th
	4,600,000	6,400	96	58	33	7	2	0	17th
	4,850,000	7,400	92	65	27	7	1	0	21st
3	4,200,000	8,200	85	57	38	3	1	1	12th
	4,200,000	6,400	90	60	36	4	0	0	17th
	4,900,000	8,800	95	65	30	4	1	0	21st
4	4,650,000	9,000	90	51	44	2	3	0	12th
	4,850,000	6,800	96	54	39	3	3	1	17th
	4,500,000	7,200	90	60	35	4	1	0	21st
5	4,150,000	8,800	95	57	41	2	0	0	12th
	4,400,000	8,200	95	55	42	2	1	0	17th
	4,200,000	7,800	90	55	43	2	0	0	21st
6	5,150,000	8,000	94	55	36	6	2	1	12th
	4,850,000	7,300	95	60	33	3	2	2	17th
	4,400,000	6,800	90	56	39	5	0	0	21st

NOTE.—It was suggested that the relatively low initial haemoglobin and red blood counts obtained in the series may have been associated with the fact that the six subjects were all young recruits comparatively recently drawn from civilian life and civilian food rationing.

DATES.—The test began on 13th August, and lasted until p.m. 17th August. Blood examined on the 12th was thus on the day before the test, on the 17th, at the end of the test, and on the 21st, 96 hours after the end of the test.

Urinalysis.—This included morning and afternoon samples examined for S.G., pH, albumen, reducing bodies, bile and indicanuria. S.G. and pH throughout conformed to normal. At no time were positive tests for any abnormal constituents obtained.

B. Plymouth. (Extracts from report by M. F. Sheehan, Surgeon Lieutenant-Commander, R.N., M.O. i/c Laboratory, R.N. Hospital, Plymouth, and Surgeon Captain H. Willoughby, R.N.V.R., N.M.O.H., Plymouth Command.)

These trials were conducted during the summer months of 1946 in the R.N. Hospital, Plymouth, and fall under two headings :—

(*a*) Those conducted in the Victualling Store Issuing Room under intermittent running of the aerosol units, *i.e.* switched on at night and off by day.

(*b*) Those conducted in the Naval Health Office where the unit was run continuously day and night for 3 months.

The aerosol units were fitted in various localities in the Plymouth Command as part of the general insecticide trials that were being carried out. It should be mentioned that so far as flies were concerned they proved very effective, with particularly successful results in the Victualling Store Issuing Room.

The subjects of the experiment were all fit people at the time of the trials. Laboratory investigations were made before and after the experiments and are shown in Table VII. In the smaller room where the aerosol ran continuously there was a slight but definite odour, particularly noticeable on first entry, after the room was closed over-night (this compares with the Porton findings) and some dryness of the throat was noticed. As, however, both the complainants were smokers it cannot be ruled out that the dry throat may have been due to tobacco rather than to the aerosol.

In the case of the Victualling Room none of the personnel noticed any odour or complained of dryness of the throat or any other symptoms.

The data of the trials are as follows :—

Victualling Store Issuing Room.

Capacity	8,300 cu. ft.
Aerosol units fitted	2
Personnel under exposure ...	3
Exposure period	30/7/46 run at nights only till 4/10/46

Naval Health Office.

Capacity	2,250 cu. ft.
Aerosol units fitted	1
Personnel under exposure ...	2
Exposure period	run continuously, night and day, for three months from 12/7/46 to 12/10/46

Before the experiment started urine examination for indicanuria (potassium indoxyl sulphate) Obermayer's qualitative test, revealed that four out of the five persons had a positive reaction. All were negative at the conclusion of the period. As controls 100 routine urines were examined for potassium indoxyl sulphate and 30 per cent. gave a positive reaction. It is considered that its presence in urine has little practical significance.

TABLE VII.

Subject	R.B.C.	W.B.C.	Hgb. Per cent.	Polys. Per cent.	Lymphs. Per cent.	Monos. Per cent.	Eosins. Per cent.	Basos. Per cent.	Indican.	Date
F.E.F.	5,630,000	12,000	110	72	20	7	1	0	—	26/7/46
	5,600,000	7,600	112	67	28	3	2	0	—	4/10/46
J.S.	5,350,000	9,000	104	57	36	6	1	0	+	26/7/46
	4,960,000	5,600	104	40	57	2	0	1	—	18/10/46
L.R.	4,600,000	6,200	92	61	31	7	1	0	+	27/7/46
	4,050,000	6,800	90	65	28	5	2	0	—	7/10/46
H.M.W.	4,420,000	7,600	94	50	44	5	1	0	+	6/7/46
	4,880,000	7,000	102	55	35	8	2	0	—	1/10/46
H.J.B.	5,000,000	10,400	96	36	55	8	1	0	+	13/6/46
	4,810,000	7,400	100	42	51	5	2	0	—	7/10/46

Haemoglobin estimation—Sahli. 100 per cent. = 14 grms. Hgb. per 100 ccs.

TOXICITY TO ANIMALS.

Porton.

The tests on animals were undertaken at Porton in order to follow up the results obtained from the initial test on human subjects. These results suggested that a more intense and prolonged exposure to such an atmosphere should be carried out, using small experimental animals, in order to obtain an indication of any potential hazard from chronic inhalation of such a DDT aerosol. This exposure was arbitrarily decided to continue for 28 days, with the volatilisation of DDT continuing throughout the period.

Test Data : a small greenhouse of 215 cu. ft. capacity was used as the test chamber. Minor ventilation was by a single roof louvre ; a single door, two shelves for animal cages and electricity supply for the heating and aerosol unit comprised the fittings. Construction was of three-quarter glass and for protection against direct insolation on clear sunny days a tarpaulin covered the roof and upper sides.

The aerosol unit was installed about 20 inches from the glass roof and 12 inches from the side, on an upright wooden support. It was charged with technical grade DDT (80 per cent. p.p. isomer content) and, once started, operated continuously for the 28 days.

Animals : Initially these were 28 guinea pigs, 20 rats (white) and 8 rabbits (hybrid). The rats and rabbits completed the whole test period but the guinea pigs developed a very infectious gastro-intestinal condition, believed to be a Gaertner infection, early in the period, and as this affected a similar proportion of all guinea pigs retained on the station, this species was deleted from the tests.

Conditions : The rats were caged in groups of five, the rabbits in pairs. Diet was normal during the test period and feeding was carried out once daily on most days ; in order to prevent over-ventilation of the chamber by entry, animals were weighed at first daily, and later in the test, every third day. They were sacrificed on the 28th day of DDT exposure and examined for pathological and histological changes.

Temperature and humidity : Within the test chamber these were determined twice daily by wet and dry bulb thermometer. Heating of the chamber proved necessary almost every night, in order to permit survival of the mosquitos being concurrently exposed.

$$\left.\begin{array}{l}\text{Average max. temp. } 80{\cdot}3^\circ\text{F.}\\ \quad\text{,, min. ,, } 60{\cdot}3^\circ\text{F.}\end{array}\right\}\text{approx.}$$

The test was carried out in two phases :—

1. Control Phase : 11th September to 23rd September—11 days control inhabitation of the test chamber without DDT to study suitability of environment and animal condition.
2. Exposure Phase : 23rd September to 21st October—28 days exposure to same environment with the additional presence of the DDT evolved from one electrical volatilisor.

The report emphasizes that this test for chronic toxicity of the DDT aerosol was necessarily restricted by current higher priority items, to a straightforward chronic exposure to determine if DDT poisoning occurred. Chemical analysis of DDT in the atmosphere or biochemical investigations on the amount of DDT absorbed or excreted etc., were not included. This was to be added in the event of a positive toxicity being established.

Results.

There were no symptoms of DDT poisoning during the entire exposure period ; animal behaviour was completely normal throughout, food consumption, alertness, muscular activity, etc., entirely as in normal animals.

Mortality during the test period was of one rabbit only, and this was due to natural causes, believed to be caused by fighting with a cage mate.

Pathological examination of the 20 surviving rats and 7 guinea pigs was carried out after sacrificing by chloroform/ether inhalation on 21st October.

All rats were found to be in excellent condition, their fat deposits and general visceral appearances being of a high standard. The livers in all cases showed completely healthy normal apearances, as did the kidneys, spleen, heart and other organs. Of these 20 rats, five were selected at random for histological examination of lungs, liver and kidney. The 7 rabbits were in likewise excellent condition and there was no specific lung, liver, kidney, spleen or other organic pathology on careful naked eye examination. Of these, three were selected at random for histological examination.

Histologically no evidence could be found for the presence of even minor DDT poisoning. The livers were universally normal, with the exception of slight diffuse fatty changes in the rabbits, which had increased in body weight considerably during the test period. Similarly the kidneys showed no cellular changes characteristic of DDT. In the lungs there were occasionally scattered peribronchial leucocyte infiltration, especially among rats, but such findings are non-specific and occurred equally among rodent controls.

Summary.

It may be stated that the summation of microscopical evidence of harmful effect upon the exposed rats and rabbits as a result of their 28 days' exposure to continuous DDT aerosol as described was nil.

Conclusions of the Toxicity on continuous-phase DDT Aerosols to Man and Animals.

The instruments were tested under conditions of normal and supra-normal insecticidal control with periods of exposure varying from 3 months continuous exposure to 5 days. Under no circumstances during any of the tests were there any indications of toxicity to man or animals.

A Review of the Literature on the Toxicity of DDT to Man and Animals.

West & Campbell (1946) describe, in a most interesting prologue to their book, the history of the development of DDT from the discovery of its insecticidal powers and the taking out of patents in 1939 to the present day. Its success as an insecticide has been meteoric with a corresponding commercial glamour, and with its release to the public shortly after the ending of the Japanese war a vast literature on every aspect of its use has been created.

In the first flush of enthusiasm extravagant claims were made for DDT which, combined with its compounding and sale by unscrupulous persons who foisted many worthless products upon an eager market, resulted in the inevitable reaction, with public and press combining to denounce both its value as an insecticide and its danger to human and animal life.

It is with the latter aspect that we are concerned here, and the following summary and analysis of the literature are given in an attempt to clarify the position and to serve as a work of reference for all interested in the use of a most remarkable and important insecticide. It is realised that much information of importance is probably hidden in the literature under other titles and there is no doubt that a great deal of first rate work carried out by members of the armed forces during the war is yet to be published or perhaps even lying fallow in unsuspected files. It is to be hoped that as much of this work as possible will be released before it is too late to profit from it. This summary is confined to the toxicology of DDT as applied to man and animals; the entomological and purely pharmacological aspects have been excluded as beyond the scope of this work.

Although we are concerned primarily with the scientific or professional literature, it is necessary, in view of the importance of the subject to the general public, to consider as well some of the pronouncements that have appeared from time to time in the lay press. Much of the post-war publicity has been frankly partisan and, in view of the paucity of published facts, necessarily uninformed and inaccurate, while a considerable amount of arrant nonsense has been talked and written. This, unfortunately, has not been confined to laymen, and persons who should have known better have been guilty of inaccurate and misleading statements. The following two examples are perhaps highlights for sheer imbecility and misleading pseudo-scientific statements respectively :—

Stage (1946) quotes the following story which apparently appeared in a southern Florida newspaper on 5th November, 1945. "*Cavalier Star Ill, Blamed on DDT Spray*. DDT may stop insect breeding but the powerful spray is hatching a nest of trouble for Ponce de Leon's football team. For a doctor's diagnosis has cited 'allergy to DDT' the cause for the illness that kept high-scoring Dave McDonald sidelined for the Cavalier's last four games. Dave's wearing a gauze mask now to try to keep free of the stuff." A colleague of Dr. Stage, Fred H. Stutz, stated : "I talked to the doctor, one of four that seem to have treated the boy the greater part of the time he was indisposed. He advised me that DDT was not thought of or mentioned in connection with his disability. The boy, apparently, had a congestion in the head and inflammation of the sinuses. No doctor said DDT had anything to do with his condition. The father stated there was no proof or evidence that DDT was involved" yet according to the news article the coach was convinced that "if the city decides to spray again before Thursday we're sunk." As Stage drily comments "A new and interesting alibi for a football team's failure, but a fallacy as to the effects of DDT."

Yarrow (1946) in a letter to the *Evening Standard* states, *inter alia*, " . . . deposit left by liquid solutions originally containing not more than 0·5 per cent. of DDT is sufficient for an efficient residual effect . . . It has been established biologically that DDT is effective in liquid preparations in as low a concentration as 0·003 per cent. . . . Very important aspect of DDT in liquid preparations is that they are toxic to human beings if put up in kerosene solution containing more than 1 per cent. of DDT." Comment is unnecessary.

The condensed data on the exposure of man and animals to DDT are presented in Table VIII. The table is divided into two sections :—

(1) Cases where the results of experimental and operational work have been non-toxic or only slightly toxic.

(2) Cases where the results have indicated definite toxic and/or lethal effects.

The table is classified, in both sections, under animal headings with sub-groupings according to the method of exposure, thus :—

(A) Aerosols and sprays.
(B) Skin (direct spraying of subject, inunction, dusting, washing, etc.)
(C) Oral.
(D) Inoculation.

TABLE VIII.

Condensed Data on Exposure of Man and Animals to DDT.

Section 1: Cases, Experimental and Operational, where no harmful Results have occurred.

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Crayfish.*	
Lackey & Steinle, 1945	In various carriers. Gas condensate liquor, kerosene, alcohol and variety of commercial detergents	0·1 lb./acre (0·3 p.p.m.) in first 3 ins. of water 20 p.p.m.	In open water unaffected if average depth of water more than 3 ins. In laboratory large individuals survived while smaller ones died.
		Fish.	
Wasicky & Unti, 1944	DDT	1 gm./20,000 litres	In aquarium; all but 3 lived normally for 60 days.
Eide, Deonier & Burrell, 1945	DDT	At mosquito larva control dosages	Mosquito control: harmless to *Gambusia*
	20 per cent. talc	1 lb./acre dusting 5 lb./ „ „	Harmless to fish.
Ginsburg, 1945 ...	2 per cent. in ethyl alcohol	1 : 20,000,000 water	Experimental on goldfish *Carassius auratus*. None dead within 6 days.
Lackey & Steinle, 1945	Various carriers: Gas condensate liquor, kerosene, alcohol and variety of commercial detergents	0·1 lb./acre (0·3 p.p.m.) in first 3 ins. of water	Fish in general unaffected if average depth over 3 ins.
		Batrachians.	
Wasicky & Unti, 1944	DDT	Water in vivarium treated 0·2 gm. DDT conc. 1/12,000 for 3 hours daily for 4 days	Batrachians.
Eide, Deonier & Burrell, 1945	DDT powder 20 per cent. in talc	Dusted at 1 lb./acre and at 5 lbs./acre	Field test: no effect on frogs.
Lackey & Steinle, 1945	Variety of carriers: Gas condensate liquor kerosene, alcohol and variety commercial detergents	0·1 lb./acre (0·3 p.p.m.) in first 3 ins. of water	Tadpoles unaffected if average depth greater than 3 ins.

* Crayfish, Fish and Batrachians, being aquatic cases, cannot be classified according to method of exposure.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Domestic Fowl.			
B) Telford, 1944	4 per cent, in pyrophyllite	Dust applied in shaker average 6·3 gm./bird	Control of chicken lice *Eomenacanthus stramineus, Menopon gallinae* on 2-year-old hens.
(C) Lepage & Giannotti, 1944	Gessarol A (3 per cent. DDT)	Oral. 100 mg. daily for 40 days	Month-old chicks.
	Gessarol P (3 per cent. DDT)	Millet treated 12 gms./60 kilo bag for 30 days	Hens.
	Pure DDT in castor oil		Hens.
Decker, 1946	DDT	Mash containing 111 p.p.m. for ten week feeding period	Well and healthy at end of period.
Decker, 1946 (Cory in discussion)	DDT	200–400 mg. in capsules	Many times normal control doses for lice. All levels within toxic and sub-toxic but not lethal ranges.
	DDT	50 mg. in capsules daily for 10, 20, 30 days	Presence of cumulative effects.
	DDT	As above, but mixed with feed	No effects.
D) Wasicky & Unti, 1944	DDT	Intravenous in fractional doses 0·2 gm. during 5 days. Oral. Twice as much for same period	1,000 gm. hen.
Pigeons.			
(C) Lent & Oliveira, 1944	Pure DDT in gelatinous capsules	Oral. 0·07–0·4 gm. to pigeons about 350 gms.	Experimental control of *Rhodnius prolixus* which were allowed to engorge on pigeons.
Wasicky & Unti, 1944	DDT	Oral. 36–50 mgs.	2 birds.
(D) Wasicky & Unti, 1944	DDT	Intravenous. 24, 34, 41 mgs. Intramuscular. 24, 34 mgs.	2 birds 2 birds } All lived normally in cage for 3 months, then killed.
Guinea Pigs.			
(A) Cameron & Burgess, 1945	15 gms./30 cc. Acetone	Thick mists, 1 : 3,000 in 1 cu. metre chamber by air driven mechanical sprayer, 2 hours daily, 6 times	

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
	Guinea Pigs—*continued.*		
A) Neal and others, 1944a	DDT 1 per cent. Sesame oil 6 per cent. Freon 93 per cent.	Aerosol: concs. of 6·22 and 12·44 mg./litre of air in static atmosphere	
Neal, Von Oettingen, Dunn & Sharpless, 1945	3 per cent. in primary solvent and secondary oil solvent with small per cent. pyrethrins	Aerosol: 45 mins. expr. to initial concs. of 26·4–32·9 mg./litre of air	No effect on adults.
B) Cameron & Burgess, 1945	Geigy DDT ... 15 Toluene ... 30 Hexamethylene tetramine ... 15 Sulphonated castor oil ... 36 Ammonia ... 4 diluted 1 : 100 water	Emulsions. 14 daily applications 5 cc./kg. (7·5 mg./kg.) to skin.	
	Pymulso concentrates: Mark X. 1 per cent. w/v DDT, 0·2 per cent. pyrethrins, 1 : 10 water	12 daily doses 5 cc./kg. to skin	
	Mark XI. 2 per cent. DDT, 0·4 per cent., pyrethrins, 1 : 20 water	Ditto	
	Special: 10 per cent. DDT, 0·4 per cent. pyrethrins, 1 : 20 water	15 doses, ditto	
	5 per cent. in pyrophyllite	200 mg./kg. (10 mg./kg. DDT) on shaved skin of back	
Wasicky & Unti, 1944	5 per cent. aqueous suspension	Compress of cotton soaked in suspension applied to skin and conjunctiva	No irritation 24 hours later.
	5 per cent. in vaseline	Ditto	Ditto
	2 per cent. solution	G. pig bathed in solution	Ditto
	Mice.		
A) Neal and others, 1944a	DDT 1 per cent. Sesame oil 6 per cent. Freon 93 per cent.	Conc. of 12·22 mg./litre air for 45 mins.	8 mice under oilcloth.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
	Mice—*continued.*		
(A) Neal and others, 1944a—*continued*	DDT 0·5 per cent. Sesame oil 6·0 per cent. Freon 93·5 per cent.	6·1 mg./litre air for 45 mins.	Mice show no symptoms only if sesame is not above 6 per cent.; at 9 per cent. toxic effects are apparent.
	DDT 5 per cent. Cyclohexanone 10 per cent. Freon 85 per cent.	Daily exposure to intermittent conc. of 0·183 mg./litre air for 45 mins. 5 days/week for 5 weeks	No toxic effects.
(C) Schmid, 1943	DDT	500 mg./kg. for 10 days orally	
Wasicky & Unti, 1944	DDT	Orally, 0·015 gm. in doses of 0·003 gm. for 5 consecutive days	2 white mice of 10 gms. each, depressed and remained in their cages from 6th day.
(D) Ditto	DDT	Intramuscular, ditto	2 white mice of 10 gms. each.
	Rats.		
(A) Neal and others, 1944a	DDT 1 per cent. Sesame oil 6 per cent. Freon 93 per cent.	12·44 mg./litre air in still air for 45 mins.	
	DDT 5 per cent. Dibutylphthallate 10 per cent. Freon 85 per cent.	54·4 mg./litre air	10 rats, reaction but complete recovery.
Ditto 1945	DDT, 3 per cent. in primary solvent and secondary oil solvent with small per cent. pyrethrins Ditto but with 2·5 per cent. DDT	Initial concs. of 26·4–32·9 mg./litre air for 45 mins.	Fatal to young rats but not to adults.
Cameron & Burgess, 1945	15 gm. in 30 cc. acetone	1 : 3,000 thick mists in 1 cu. metre chamber by air driven mechanical sprayer 2 hours daily, 6 times	

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Rats—*continued.*			
C.D.E.S. Porton (unpublished)	Electrical volatilisation of commercial grade DDT (80 per cent. p.p. isomer) to form a continuous phase aerosol.	One unit of average output of 0·75 gm./day of average particle size 0·5–5·0μ situated in greenhouse of 215 cu. ft., 20 ins. from roof and 12 ins. from side. Operating continuously for 28 days. Estimated total output approx. 21 gm.	20 rats. No signs whatever of DDT intoxication or pathological changes due to absorption of DDT.
Cameron & Burgess, 1945	1 per cent. with ethyl alcohol, acetone and ether as solvents	Daily skin applications of 2 hours up to 12	
	Geigy DDT ... 15 Toluene ... 30 Hexamethylene - tetramine ... 15 Sulphonated castor oil ... 30 Ammonia ... 4 diluted 1 : 100 water	14 daily skin applications of 5 cc./kg. (7·5 mg./kg.)	
	Pymulso concentrates: Mark X. 1 per cent. w/v DDT, 0·2 per cent. pyrethrins, 1 : 10 water	12 daily doses 5 cc./kg. (applied to skin)	
	Mark XI. 2 per cent. w/v DDT, 0·4 per cent. pyrethrins, 1 : 20 water	12 daily doses of 5 cc./kg. applied to skin	
	Special: 10 per cent. w/v DDT, 0·4 per cent. pyrethrins 1 : 20 water	15 doses, ditto	
	5 per cent. in pyrophyllite	500 mg./kg. (10 mg./kg. DDT) of powder applied to skin	
Draize & others, 1944	DDT	Applied to diets at 100, 200 and 400 p.p.m.	400 p.p.m. showed nervous symptoms, 200 p.p.m. slight effect on growth.
Smith & Stohlman, 1944	DDT in olive oil	50 and 100 mg./kg.—oral	Hyperexcitability and tremors but no mortality.
Wasicky & Unti, 1944	DDT in colloidal solution	1·2 gm. in doses of 0·2 gm. during 6 consecutive days	

TABLE VIII, Sect. 1—*continued.*

	Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
	Rats—*continued.*			
(C)	Cameron & Burgess, 1945	5 per cent. suspension in liquid paraffin and tragacanth	50 mg./kg. by stomach tube 30 doses. Total 1·59 kg.	
	Rabbits.			
(A)	Neal and others, 1944a	1 per cent. DDT in Deobase	Heavy mist 48 mins. daily for 4 weeks	No signs of poisoning but some irritation of mucous membranes of eyes and upper respiratory tracts and skin caused by solvent.
	Cameron & Burgess, 1945	15 cc. in 30 cc. acetone	Heavy mist. 1 : 3,000 in cu. metre chamber by air driven mechanical sprayer 2 hours daily, 6 times	
	C.D.E.S. Porton (unpublished)	Electrical volatilisation of commercial grade DDT (80 per cent. p.p. isomer) to form a continuous phase aerosol	One unit of average output of 0·75 gm./day of average particle size 0·5–5·0μ situated in greenhouse of 215 cu. ft. 20 ins. from roof and 12 ins. from side. Operating continuously for 28 days. Estimated total output approx. 21·0 gm.	8 rabbits: 1 died from injuries believed due to fighting with cage-mate. Remainder showed no signs of DDT intoxication or pathological changes due to absorption of DDT.
(B)	Cameron & Burgess, 1945	Geigy DDT 15 parts Toluene 30 parts Hexamethylenetetramine 15 parts Sulphonated castor oil 36 parts Ammonia 4 parts diluted 1 : 100 water.	Emulsions 14 daily skin applications 5 cc./kg. (7·5 mg./kg.)	
		Pymulso concentrates: Mark X. 1 per cent. w/v DDT, 0·2 per cent. pyrethrins, 1 : 10 water	12 daily doses applied to skin 5 cc./kg. (7·5 mg./kg.)	
		Mark XI. 2 per cent. w/v DDT, 0·4 per cent. pyrethrins, 1 : 20 water	Ditto	
		Special: 10 per cent. DDT, 0·4 per cent. pyrethrins, 1 : 20 water	15 doses of 5 cc./kg. applied to skin	

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Rabbits—*continued.*			
B) Cameron & Burgess, 1945—*continued.*	0·5 per cent. in benzene	Impregnated cloth allowed to dry and strapped to skin (shaved) for 7 days. Doses of 10, 25 and 100 mg./kg.	5 rabbits.
Draize and others, 1944	In 30 per cent. solution of dimethylphthallate	Single doses of 3, 9, 6, and 9·4 ml./kg. to intact and abraded skin	All animals showed toxic signs but none died.
Elmes, 1945	5 per cent. in acetone	Swabbing of ears, feet dipped in solvent	Treatment of Psoroptic and Sarcoptic mange. Five with former and six with latter. Marked clinical improvement of treated animals.
Cameron & Burgess, 1945	5 per cent. in pyrophyllite Plain powder	200 mg./kg. (10 mg./kg. DDT) 50 mg./kg. on shaved skin of backs. 16 daily doses of two hours.	
Schmid, 1943	5 per cent. solution	Applied to cornea	1 rabbit only. No injurious phenomena during 7 days' observation.
Wasicky & Unti, 1944	0·5 per cent. in aqueous suspension	Applied to eyeballs and conjunctiva	No irritation.
(C) Schmid, 1943	DDT	100 mg./kg. orally	
Lindquist, Knipling, Jones and Madden, 1944	DDT	228–537 mg./kg. orally	Control of *Cimex lectularius* and *C. hemipterus* feeding on rabbits. One died at 537 mg. From 80 per cent. to 100 per cent. mortality of bugs.
Lepage & Giannotti, 1944	Pure DDT	2·64 gms. orally over 17 days	Gained 10 per cent. in weight.
Smith & Stohlman, 1944	DDT in olive oil	50 and 100 mg./kg. orally	5 and 6 rabbits respectively, hyperexcitability but no deaths.
Cameron & Burgess, 1945	5 per cent. emulsion in liquid paraffin and tragacanth	Repeated doses by stomach tube of 50 mg./kg. up to 7 doses	None died at 7 doses.
(D) Philips & Gilman, 1946	DDT	25 mg./kg. intravenous 35 mg./kg. ,,	

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Cats.			
(C) Smith & Stohlman, 1944	DDT	100 mg./kg. orally	1 cat. Results from cats very varied.
Wasicky & Unti, 1944	DDT	315–480 mg. ditto	2 cats.
Philips & Gilman, 1946	10 per cent. in pea nut oil	200 mg./kg. ditto 450 mg./kg. ditto	Very erratic as cats died from intermediate doses.
(D) Wasicky & Unti, 1944	DDT	48–72 mg. intramuscular	2 cats.
Philips & Gilman, 1946	As emulsion	25 mg./kg. intravenous	
Dogs.			
(A) Gouck & Smith, 1944	20 per cent. solution benzyl benzoate	By atomiser	Successful control of ticks *Rhipicephalus sanguineus* and *Amblyomma americanum.*
Neal and others, 1944a	DDT 1 per cent. Sesame oil 6 per cent. Freon 93 per cent.	12·22 mg./litre air for 45 mins. in static air	2 puppies.
	Ditto	Daily exposure for 45 mins. to concentration of 12·2 mg./litre air on 2 successive days in one week and 4 successive days the following week	2 puppies, gained weight.
	DDT 5 per cent. Dibutylphthallate 10 per cent. Freon 85 per cent.	54·4 mg./litre air	2 puppies. Reaction but recovered completely.
	DDT 5 per cent. Cyclohexanone 10 per cent. Freon 85 per cent.	Ditto	Dogs narcotised but recovered.
Neal and others, 1945	3 per cent. in primary solvent and secondary oil solvent with small per cent. pyrethrins As above but with 2·5 per cent. DDT	Repeated single massive doses of mean initial conc. of 33 mg./litre air DDT	Minor irritation and narcotic action due to primary solvent.
(B) Gouck & Smith, 1944	DDT 5 parts Benzene 6·5 parts Commercial emulsifier 1·25 parts Water 87·5 parts	Washing	Control of ticks *Rhipicephalus sanguineus, Amblyomma americana.* Complete kill with long and short-haired dogs.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Dogs.—*continued.*			
B) Gouck & Smith, 1944—*continued.*	5 per cent. emulsion in "B 1956" 5 per cent. emulsion in "Dreft" 5 per cent. emulsion in "Vatsol"	Washing	Successful control of ticks.
Stage, 1946	5 per cent. benzene emulsion 1 per cent. solution pine oil 1 per cent. water dispersible suspension	Part of total of more than 200 treatments	45 dogs, no ill effects. Flea and dog tick control.
Gouck & Smith, 1944	5 per cent. in pyrophyllite	Dusted on	Failed to kill ticks.
Lindquist, Madden & Knipling, 1944	5 per cent. in pyrophyllite	10 gms. to medium sized dog rubbed into hair	Dog flea control, *Ctenocephalides canis*, *C. felis*, *Echidnophaga gallinacea*. 11 dogs, very effective. Res. tox. 7 days.
Neal and others, 1944a	10 per cent. DDT, 90 per cent. Pyrax ABB	Daily exposures for 3 hours over 4 weeks to concs. 12·48 mg. DDT per cu. metre.	
Wasicky & Unti, 1944	5 per cent. powder	Applied to skin	
Stage, 1946	10 per cent. in pyrophyllite 10 per cent. in talc	Part of more than 200 treatments	Part of experiment on 45 dogs.
Annand, 1944	0·5 per cent.	?	Stated to be invaluable in protecting dogs from *Stomoxys calcitrans*.
(C) Draize and others, 1944	DDT in gelatin capsules	10 and 50 mg./kg. per day orally 6 days a week for 50 weeks	One dog at 50 level after losing weight for long time died on 34th week.
Neal and others, 1944	In capsules In powder	100 mg./kg. insufflated	No symptoms at low doses.
Wasicky & Unti, 1944	5 per cent. DDT mixture	5 gms. to dogs of 5–6 kg. weight for 31 days	2 dogs.
Vaz, Pereira & Malheiro, 1945	10 per cent. oily solutions	Daily dose of 100 mg./kg. until symptoms appeared; average quantity taken 24·8 gms.	6 dogs, all but one in good health a month later.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Dogs—*continued.*			
(C) McNamara, Bing & Hopkins, 1946	DDT	Daily doses of 150–300 mg./kg.	Severe symptoms but no deaths. Recovery after DDT discontinued.
(D) Philips & Gilman, 1946	DDT	35, 50, 60 mg./kg. intravenously	
Pigs.			
(C) Decker, 1946	DDT	Corn containing DDT far in excess of possible residues for period of several months	No symptoms and no damage to muscles, nerves, kidneys or liver.
Decker, 1946, (reporting for Lardy in discussion)	DDT	500 p.p.m. in diet orally since time of weaning to halfway through gestation	One female, no evidence of toxicity.
Goats and Sheep.			
(B) Schmid, 1943	2 per cent. emulsion	Dipping	Nursing sheep and lambs, no reaction.
Babcock, 1944	0·3 and 0·6 per cent. in water emulsion	Dipping	Angora goats, control of lice: *Holokartikos crassipes*, *Damalinia* (*T.*) *caprae*, *Linognathus stenopsis* and Trichodectids.
Anon., 1944b	DDT emulsion	Moderate doses	Doses given to pregnant ewes; normal lambs born. Several hundred goats dipped, eyelids examined, no damage.
Telford & Guthrie, 1946	5 per cent. and 10 per cent. emulsion. Formula presumably as for cattle	Excessive quantities. Young goat 35 lb. wt. 50 cc. spray applied 11 times weekly for 12 weeks;	No signs of macroscopical changes or DDT poisoning on dissection.
		75 cc. of 10 per cent. spray 6 times week for 5 weeks	Ditto.
		Goat of 139 lbs., 150 cc. applied 11 times weekly for 8 weeks.	Ditto.
		Pregnant♀, 150 cc. of 10 per cent. applied 11 times weekly for 3 weeks	2 kids born, both died within 48 hours with tremors and convulsions. Other kids put to suck, with spraying continuing, no ill effects.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Goats and Sheep—*continued.*	
(C) Schmid, 1943	1 per cent. solution	Grass sprayed for one week	6 sheep.
Cox, 1944	10 per cent. dust	40 lbs. per acre	3 ewe sheep grazed after 48 hours showed extreme symptoms after 78 hours, but recovered after rain and wind had removed dust.
		Same experiment repeated one month later	Same results.
West & Campbell, 1944	Suspension of "Gesarol" in water	10–100 lbs./100 Imperial gallons on green fodder	Sheep: increase in weight in all tests. No symptoms.
Stage, 1946	DDT	2 lbs. and 7½ lbs. per acre sprayed or dusted	2 goats, 2 sheep.
Decker, 1946	DDT	In silage for several months	Normal health and appetite.
Decker, 1946 (reporting for Lardy in discussion)	DDT	Pea vines made into silage with 1 lb. DDT added per ton at time of preparation equals application of 6 lbs./acre	No toxic symptoms.
Cox, 1944	DDT	2 gm. for 11 days orally 4 gm. for 40 days orally 8 gm. for 14 days orally	1 ewe sheep.
Telford & Guthrie, 1945	DDT	1 gm. per 8–9 lbs. body weight for 21–26 days	Goats: lactation generally ceased between 21st and 28th day. Some animals showed no toxic symptoms and all recovered, yet their milk produced symptoms in rats.
Orr & Mott, 1945	As dry powder and water suspension	100 mg/kg. daily for 1st week	4 sheep.
		150 mg./kg. daily for 2nd week	3 sheep.
		200 mg./kg. daily for 3rd week	
Neal & von Oettingen, 1946	DDT in olive oil	Single dose of 1,000 mg./kg. by stomach tube	Temporary nervousness and tremors, a dose of this magnitude proves fatal only if it has been administered repeatedly.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Cattle.			
(A) Wells, 1944	DDT 5 per cent. Sesame oil or acetone 10 per cent. Freon 113 20 per cent. Methyl chloride 65 per cent.	Applied as aerosol	Control of *Lyperosia irritans* Res. tox. 4 days.
(B) Ditto	DDT 100 gms. Benzene 140 ml. Kerosene 140 ml. Triton, N.E. 8 gm. Water to make 1 litre	Emulsions applied by power sprayer	90 cattle sprayed in 1 hour with one nozzle. Res. tox. 1 week.
	DDT 100 gm. Benzene 140 ml. Dibutyl Phthallate 140 ml. Triton H.E. 12 gm. Water to make 1 litre	Ditto	No ill effects.
Australia, 1944	2 per cent. alcoholic emulsion	Spraying	Control of *Boophilus annulatus microplus, Lyperosia exigua*. Res. tox. 2–13·5 days.
	4 per cent. kerosene solution		Herds free for 2–3 weeks. Kerosene injurious to skin of cows if applied too heavily.
Munro & Knapp, 1945	10 per cent. in pyrophyllite	Dusting	Control of *Haematopinus eurysternus* and *Bovicola bovis* on 3 yearling steers and 3 2-year old heifers — complete control of lice in 5 weeks.
Telford & Guthrie, 1946	DDT 10 parts Velsicol AR 60 44 parts Water 40 parts Pine oil 6 parts. Plus small amount of either Triton X100 or Vatsol OS	100 cc. at each application by hand sprayer twice daily for 5½ days a week for 12 weeks	2 purebred Holsteins in similar periods of lactation. Milk yields normal, no weight change. Gave birth to normal young 3½ and 7 months after close of experiment.
Rude & Smith, 1944	2 per cent. and 5 per cent. in non-drying adhesive	Applied to inside and outside of ears	Control of *Amblyomma maculatum* and *Otobius megnini*. 416 cattle treated.
(C) Schmid, 1943	1 per cent. solution	Sprayed on grass and fed to cattle for one week	2 cows.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Cattle—*continued.*			
(C) West & Campbell, 1944	Suspension of "Gesarol" in water	10–100 lbs. per 100 imperial gallons on green fodder	Volume of milk, quality, process of calving, soundness and growth of calves all normal.
Orr & Mott, 1945	DDT	100 mg./kg. for 1st week 150 mg./kg. for 2nd week 200 mg./kg. for 3rd week	4 cows.
Stage, 1946	DDT	1½ oz. daily for 6 days By grazing 2 lb./acre By grazing 7½ lb./acre	900 lbs. cow showed no symptoms. 2 cattle gave signs of stiffness after the 6th day at 7½ lb. Recovered when removed to clean pasture.
Decker, 1946	5 per cent. dust	Fed 40 lbs. daily for 3 months on silage dusted 3 times at rate of 20 lbs./acre	Steers, normal appetite and health, no tissue damage.
	DDT	Treated silage for several months	Normal health and appetite.
Decker, 1946 (reporting for Lardy in discussion)	DDT	Pea vines made into silage with 1 lb. DDT/ton added when made, equals application of 6 lb./acre. Estimated max. dose 9 gms./cow/day	No toxic symptoms, milk fed to rats without ill effects on growth or behaviour.
Annand, 1944	0·5 per cent.	Method unknown	
Horses.			
(B) Blakeslee, 1944	In various solvents: best formula DDT 2 gms. Indalone 100 cc.	4 oz. per animal twice daily up to 35 applications of spray	Res. tox. on animal for a few days.
(C) Orr & Mott, 1945	DDT	100 mg./kg. for 1st week orally 150 mg./kg. for 2nd week orally 200 mg./kg. for 3rd week orally	2 horses.
Stage, 1946	DDT	2 lbs./acre 7½ lbs./acre dusting and spraying pastures	2 grazing horses.
Annand, 1944	0·5 per cent.		Stated to be effective in protecting horses from *Stomoxys calcitrans*.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Monkeys.	
(A) Neal and others, 1944a	DDT 5 per cent. Cyclohexanone 10 per cent. Freon 85 per cent.	Chronic exposure of 0·183 mg./kg. litre air every 15 mins. for period of 45 mins 5 days a week for 5 weeks	
	Ditto	As above, but for 3 periods daily for 4 weeks.	
Neal, von Oettingen, Dunn & Sharpless, 1945	3 per cent. in primary solvent and secondary solvent with small percent. pyrethrins. Also with 2·5 per cent. DDT	Exposure for 45 mins. daily on 119 occasions over period of 2 weeks to median initial conc. of 33·3 mg./litre air	Rhesus monkeys.
(C) Hill & Robinson, 1945	5 per cent. in kerosene	15 cc. per body weight of 3·2 kg. orally	Recovery in 2 hours.
(D) Philips & Gilman, 1946	DDT emulsion	40 mg/kg. intravenous	One monkey only used.
		Man.	
(A) Neal and others, 1944a	DDT 5 per cent. Cyclohexanone 10 per cent. Freon 85 per cent.	Intermittent exposure of 0·035 mg./litre air (1 gm./1,000 cu.. ft.) for 1 hour daily for 6 days Exposure intensified by introducing 0·4 gm. of aerosol at intervals of 5 instead of 15 mins.	2 men aged 42 and 54 years. One of these two men went on to further oral experiments (see under).
Neal, von Oettingen, Dunn & Sharpless, 1945	DDT	Relatively heavy exposure to DDT over period of 14 weeks	Four individuals thorough clinical and laboratory examination revealed no injurious effects.
C.D.E.S., Porton. (Unpublished)	Electrical volatilisation of commercial grade DDT (80 per cent. pp. isomer) producing a continuous phase aerosol	Apparatus sited on wall of 4,000 cu. ft. test room, 7 ft. from ground and 2 ft. below ceiling. Average output of unit 0·75 gm./day, average particle size 0·5–5·0μ. Three units in continuous operation for 9 days, 4 days as a build-up before exposure of subjects and 5 days	Six service (R.N.) volunteers, all medically fit and clinically examined before and during test. In order to prevent psychogenic complaints full amenities provided, games, books, radio, etc. Results entirely negative. No complaints from men: blood counts and urine analysis negative

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Man—*continued.*			
		during exposure. Exposure from 9.30 to 12.30 and from 2.0 to 4.30 p.m. daily for 5 days. Total exposure time of 27½ hours. Average total output during exposure period 2·25 gm. DDT approx. Average total output during 9 days was 20·25 gm. DDT. approx.	Eyes, nose, throat, skin, senses and general condition normal.
Willoughby & Sheehan, R.N. Hospital, Plymouth (Unpublished)	As above	2 units installed in victualling issuing room. 8,300 cu. ft. Operating every night but shut off in day for three months 1 unit in Naval Health office, 2,250 cu. ft. operating continuously for 3 months	3 personnel working under normal conditions. No complaints of any kind. Negative haematological and urinalysis findings. 2 subjects: 2 complained of slight sore throat, but both smokers. Otherwise negative results
(A B) Neal and others, 1944	Solution of 5 per cent. (sometimes 7 per cent.) DDT in cyclohexanone and kerosene 12·5 gals. kerosene to 1 gal. solution of DDT in cyclohexanone 25 per cent. DDT/Talc. Aerosol formula: Freon, DDT and Oil	Two men were continuously engaged from October 1943 to January 1944 in spraying and dusting barracks, mess halls and other living quarters, mostly using the solution but occasionally dusting. Average consumption of solution was 14 gals. a day, each man spraying half the time. Power spray used. Since January only two days a month averaged. Two sets of fatigue clothes used and underclothes were changed 2 or 3 times a week. August 1943 to January 1944, one man spent 90 per cent. of time with aerosols, 10 per cent. with sprays and since that time to present (9 months in all) 20 per cent. aerosols and 80 per cent. sprays. For 9 months was exposed 5 days a week, mostly experimental work. Had 1 hour exposures daily to inhalations of concentrations varying from 20 to 100 mg./1,000 cu. ft. and hands frequently soiled. None of the three men complained of any results attributable to DDT and the authors state "The results of the examinations of these three men can be summarised in the statement that after carefully evaluating all the evidence accumulated from each case we are led to the conclusion that none of them present definite findings that can be attributed to the toxic action of DDT.	
B) Wasicky & Unti, 1944	Concentrated solution	Cotton wicks soaked in solution held with plaster on inner arm	3 men, no reactions after 24 hours.
	2 per cent. ointment	Rubbing of groin	Control of *Phthirus pubis* on group of people.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Man—*continued.*	
(B) Anon., 1945a; see also Dangerfield, 1946	DDT used in all expts. was from one package: sample analysed at 59 per cent. pp : isomer Following preparations tried :— (A) 5 per cent. in 2nd grade kerosene (B) 5 per cent. in 3rd grade kerosene (C) 5 per cent. in 2nd grade kerosene and lubricating oil mixture (D) No. 1 Anti-gas lab. standard DDT concentrate :— Turpentine 33·4 per cent. Toluene 33·4 per cent. Rectified spirit 4·9 per cent. Soft soap 3·0 per cent. Water 0·3 per cent. Commercial DDT 25·0 per cent. (E) 12·5 per cent. in oil/water emulsion prepared by 1 : 1 water dilution from (D) (F) 5 per cent. in oil/water emulsion from (D) 1 : 4 (G) 20 per cent. DDT in toluene (H) Sat. soln. in alcohol (2 per cent.) (I) 20 per cent. in acetone (J) 5 per cent. in castor oil (K) 5 per cent. in arachis or ground nut oil	Up to 10 applications a day for 3 days. Hands and forearm washed at 12.45 hours and 16.45 hours for taking meals. Average time of contact with DDT 5 hours a day. Hands and forearm most frequent sites, knees also used. Liquid applied either by swabbing with cotton wool or by pouring it on to back of hand and spreading with the other. Applications made at suitable times of the day, frequency of application increased as tests became more and more severe. Amount applied each time also increased but it was found wasteful to apply more than 15–20 cc. at once. In case of solutions of kerosene and in more volatile agents solvent had evaporated before next application, but oily solutions remained on skin and subsequent applications were made on residue of previous one. In all experiments there were no toxic symptoms or signs attributable to DDT and no abnormality was found in any specimen of urine examined. 62 men exposed, 42 Indian, 20 British.	
Cameron & Burgess, 1945	1 per cent. DDT dry weight basis	Impregnated clothing	12 soldiers wore woollen vests, long woollen drawers and Angola shirts impregnated with DDT (1 per cent.) for 18–26 days, carrying out partial military duties and undergoing severe exercise to induce sweating. Physi-

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Man—*continued.*	
			cal, blood and urine investigations carried out. No symptoms and negative results. Similar conclusion on 2nd experiment with 40 men.
	DDT	Bulk impregnation during making up solutions, etc.	Six workers kept under observation for 77 days impregnating clothing, making up large vols. DDT solutions, distilling residues. Wore protective clothing, including gloves and gum boots. One man showed rise in blood calcium, but otherwise fit, rest showed no detectable changes.
Original	5 per cent. in grade 2 kerosene	Operational exposure to mist and intense soaking over 9 months during which 175 spray operations carried out in ships and shore establishments with consumption of 9,402 gallons of solution and a total spraying time of 566 hours	15 men: 14 Cingalese, 1 Tamil. Ages ranged from 21–40 years with average of 26·4.
(C) MacCormack, 1945	Commercial DDT in butter	Swallowed by author approx. 1·5 gm.	No ill effects except subcutaneous haemorrhages in pressure spots (fingers of left hand and outside of left foot) when playing golf.
Lazar, 1946	Commercial DDT in pancakes	Half - dozen pancakes made of DDT powder instead of flour	Eaten by an officer in a mess in Germany and witnessed by many of his comrades. No ill effects.
Neal, Sweeney, Spicer & von Oettingen, 1946	DDT in olive oil	11 mg./kg. Total dose of 770 mg.	The same subject had previously been exposed to aerosols (see Neal, von Oettingen *et al.* 1944) and later, in December 1944, had ingested 500 mg. DDT in olive oil with no ill effects.
Neal, 1946	5 per cent. in Deobase		Drunk by a child, but prompt action in washing out stomach resulted in recovery.

TABLE VIII, Sect. 1—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Man—*continued*.	
(A B C) Fennah, 1945	DDT. Pure 3 per cent. emulsion	During 13-month period, interupted for fortnight and month, respectively, inhaled 100 mg. of sifted pure DDT and drank water dusted with pure DDT at more than 300 mg./sq. ft. daily. At later date, for a period of a month, all food taken was sprayed when prepared and exposed in dishes with 3 per cent. emulsion until a plane surface exposed at same time had received 100 mg./sq. ft. Finally at irregular intervals of 15 days, the skin of the arms and hands was swabbed with 5 per cent. DDT emulsion and allowed to dry, giving a dosage of over 200 mg./sq. ft.	No ill effects of any kind and no symptoms associated with DDT poisoning in laboratory tests were noted.

Section 2.—Toxic and Lethal Results.

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Mollusca.	
Eide, Deonier & Burrell, 1945	2·0 ppm in sulphonated sperm oil in ethyl ether of ethylene glycol	In natural water	Snails: *Physa* sp., dead within 24 hours.
		Crayfish.	
Lackey & Steinle, 1945	In various carriers. Gas condensate liquor, kerosene, alcohol and variety of commercial detergents	1·0 ppm 0·25 ppm in water from which there is no escape	Lethal to crayfish. Some mortality with crayfish.
		Fish.	
Wasicky & Unti, 1944	DDT	1 gm./20,000 litres in aquarium	3 fish out of a number dead.

TABLE VIII, Sect. 2—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Fish—*continued.*	
Eide, Deonier & Burrell, 1945	DDT	In concs. greater than 0·1 p.p.m.	Toxic to *Carassius auratus*.
	2·0 p.p.m. in sulphonated sperm oil in ethyl ether of ethylene glycol	Natural water	Large-mouthed black bass, *Huro salmoides*, died after few hours: at end of week all fish including top minnows were dead.
Ellis, Westfall & Ellis, 1944	DDT in olive oil	Single doses incorporated in food pellets. Amounts ranging from 63–200 mg./kg.	Fed to goldfish of 6–10 grm weight. 55 per cent. toxicity within this range.
Ginsburg, 1945	Diluted kerosene emulsion. 0·1 per cent. DDT 0·2 per cent. DDT		Fatal within 3 days to the black bass, *Micropterus salmoides*. Cat fish, many dead in 2 days.
	DDT/Xylol	1 : 4,000,000	Salt-water minnows, *Fundulus heteroclitus*, 5 dead in 2 days in salt marsh test holes 2 ft. deep, 54 ft. square.
	Ethyl alcohol 2 per cent. DDT	In laboratory 1:500,000 or lower between 1 : 1,000,000 and 1 : 5,000,000 1 : 10,000,000	*Carassius auratus*, all died within 24 hours; 100 per cent. kill within 2 days. 40 per cent. dead at end of 6 days.
	Water, white kerosene, 3 per cent. DDT emulsified with sodium lauryl sulfate	1 : 2,500,000 lbs./acre 0·42 1 : 1,125,000 ,, 0·85 1 : 562,000 ,, 1·7	30 per cent. dead after 3 days, 60 per cent. dead after 6 days. 40 per cent. and 60 per cent. 60 per cent. and 80 per cent.
	Dusts: 1 and 0·1 per cent. DDT from commercial sample of 20 per cent. DDT/pyrophyllite with soapstone diluent	1 : 10,000,000 ,, 0·1 1 : 1,000,000 ,, 1·0 2 lbs./acre	10 fish in experiment. 33 per cent. kill after 6 days in this range. Average kill 83 per cent.
Lackey & Steinle, 1945	In gas condensate liquor kerosene, xylene, alcohol and variety of commercial detergents	1·0 p.p.m. surface spraying and emulsion fed into current 0·25 p.p.m. in water from which there is no escape 0·1 p.p.m.	Toxic to *Gambusia affinis*; water remains toxic for 5 days. Some mortality in fish in the field. Under lab. conditions toxic to fish in 24 hours.

TABLE VIII, Sect. 2—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Fish.—*continued.*			
Buxton, 1945	5 per cent. in dieseline	0·5 cc. of emulsion per square yard	Killed small fish in shallow pit.
Stage, 1946, quoting Cottam	DDT	Applied at ½ lb./acre as mosquito control on a peninsula several miles long and few hundred yards wide by aircraft. As 20 m.p.h. wind was blowing pilot released several times the intended dose	Three days later an estimated 100,000 small fish dead: Menhaden, mullet and Killifish were seen along entire shore line.
Batrachians.			
Lackey & Steinle, 1945	In various carriers; gas condensate liquor, kerosene, xylene, alcohol and variety of commercial detergents	Overall conc. of 0·25 ppm. in water from which no escape	Toxic to tadpoles.
		0·1 p.p.m.	Under lab. conditions toxic to tadpoles in 24 hours.
		1·0 p.p.m.	Toxic to tadpoles in field.
Ellis, Westfall & Ellis, 1944	DDT	Injection of single dose of 150 mg./kg. into dorsal lymph sac	All dead in 4–72 hours.
Reptiles.			
Eide, Deonier & Burrell, 1945	DDT sulphonated sperm oil in ethyl ether of ethylene glycol	2·0 p.p.m. in field	Small water snake dead after 1 day. Salamanders dead in one week.
Ginsburg, 1945	Kerosene emulsion diluted 0·2 per cent.	Applied to pond.	Two water snakes and one turtle dead after 2 days.
Pigeons and Chicks.			
(C) Lent & de Oliveira, 1944	Pure DDT in gelatinous capsules	0·45–0·5 gm.	Pigeons seriously affected, some fatally.
Woodard, Nelson & Calvery, 1944	DDT	0·05 per cent. in diet	Definite toxic signs in young chicks.
Guinea Pigs.			
(A) Cameron & Burgess, 1945	15 gm. in 30 cc. acetone	Nominal conc. 1:1,000 thick mist in 1 cu. metre chamber with air-driven mechanical sprayer. Exposure 2 hours, 11 times	1 guinea pig died after 9 exposures, 4 died after 10 exposures.

TABLE VIII, Sect. 2—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Guinea Pigs—*continued.*			
Draize, Nelson & Calvery, 1944	Solution of DDT in Dimethylphthallate	Inunction of 150 mg./kg./day for 30 days	Death after 30 days.
Draize and others, 1944	30 per cent. solution in Dimethylphthallate	Skin doses of 0·5, 1·0, 2·0 and 4·0 ml./kg. daily for 13 weeks	Death at all levels preceded by characteristic toxic signs.
Cameron & Burgess, 1945	10 and 1·0 per cent. kerosene solutions. 1 per cent. ethyl alcohol acetone and ether solutions	10 per cent. kerosene solution 200 mg./kg.	5 guinea pigs, all died after 12 applications.
		1 per cent. solution 50 mg./kg. daily skin application of 2 hours	5 guinea pigs, all died after 18 applications.
		1 per cent. acetone at 10 mg./kg.	2 of 5 died at 7 and 9 applications.
		1 per cent. ether at 10 mg./kg.	2 of 5 died at 7 and 8 applications.
	With ether or kerosene as solvents	L.D. 50 for single doses is 1,000 mg./kg.	L.D. 50.
Woodard, Nelson & Calvery, 1944	DDT in corn oil	Single oral doses of > 562 mg./kg. 0·1 per cent. level in diet	L.D. 50.
Domenjos, 1944	Emulsion of gum arabic	2·0 gm./kg. orally	L.D.
Draize and others, 1944	DDT in corn oil	400 mg./kg.	L.D. 50.
	DDT	Mixed in diet 500 p.p.m. equals daily intake of 40 mg./kg.	Toxicity within a few weeks.
Cameron & Burgess, 1945	In liquid paraffin	Gastric administration 400 mg./kg.	L.D. 50.
Woodard, Nelson & Calvery, 1944	DDT in corn oil	Intra-muscular, intra-peritoneal, sub-cutaneous	L.D. 50 higher than for oral administration but action irregular.
Cameron & Burgess, 1945	Liquid paraffin	900 mg./kg. sub-cutaneous	L.D. 50.
Mice.			
Neal and others, 1944a	DDT 1 per cent. Sesame oil 6 per cent. Freon 93 per cent.	12·4 mg./litre air, 45 mins. exposure	20 mice, 3 died 24 hours, 4 died 72 hours, 1 at 96 hours.

TABLE VIII, Sect. 2—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Mice—*continued.*			
(A) Neal and others, 1944a	DDT 5 per cent. Cyclohexanone 10 per cent. Freon 85 per cent.	0·183 mg./litre air every 15 mins. for 45 mins. 3 times daily, 5 days per week for 4 weeks In special experiment using above formula 30 mice used, 10 unprotected, 10 with fur collars to prevent licking of fur, 10 wrapped in gauze to prevent contamination of fur. Protected animals, 9 died of narcosis and surviving mouse showed no signs of DDT poisoning. Unprotected and to lesser extent the collared mice developed typical signs of DDT poisoning and all died after varying periods of time.	Death to mice.
	DDT 5 per cent. Dibutylphthallate 10 per cent. Freon 85 per cent.	54·4 mg./litre air	7 of 10 mice died.
Neal, von Oettingen, Dunn & Sharpless, 1945	3 per cent. DDT in primary solvent and secondary solvent with small percentage pyrethrins Also with 2·5 per cent. DDT	Initial conc. of 26·4 to 32·9 mg./litre air for 45 mins.	L.D. 50 for immature mice.
		0·11 mg./litre air exposure of 50 mins, 3 times a day, 5 days per week for 6 weeks Produced by repeated dispersal of aerosol	Definite toxic effect.
(C) Domenjoz, 1944	Gum arabic emulsion in olive oil	1·6 gm./kilo orally	Lethal doses.
		0·175 gm./kilo orally	
Woodard, Nelson & Calvery, 1944	DDT in corn oil	448 mg./kg.	L.D. 50.
	DDT	Levels in diet of 0·05 per cent.	Toxic to mice.
Draize and others, 1944	DDT in corn oil	400 mg./kg. orally,	L.D. 50.
	DDT	Levels below 500 p.p.m.	Toxic to mice.
(D) Woodard, Nelson & Calvery, 1944	DDT in corn oil	Intramuscular, intraperitoneal, subcutaneous	L.D. 50, higher than for oral (448 mg./kg.) but irregular.

TABLE VIII, Sect. 2—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Rats.	
A) Cameron & Burgess, 1945	15 gm./30 cc. acetone	Thick mists, 1 : 1,000 in 1 cu. metre chamber by air-driven mechanical sprayer 2 hours daily, 11 times	10 rats. 1 died after 4 exposures. 3 died after 5 exposures. 2 died after 6 exposures. 1 died after 8 exposures. 2 died after 9 exposures. 1 died after 10 exposures.
Neal, von Oettingen, Dunn & Sharpless, 1945	3 per cent. in primary solvent and secondary oil solvent with small percentage pyrethrins, also with 2·5 per cent. DDT	Concs. of 26·4–32·9 mg./litre air for 45 mins.	Fatal to 15 per cent. immature rats.
B) Draize, Nelson & Calvery, 1944	DDT in Dimethylphthallate	150 mg./kg./day for 30 days applied to skin	Caused death in some cases.
Draize and others, 1944	DDT in 30 per cent. solution of Dimethylphthallate	Daily doses of 0·5, 1·0, 2·0 and 4·0 mg./kg for 13 weeks applied to skin	Deaths at all levels preceded by characteristic toxic signs.
Cameron & Burgess, 1945	10 per cent. kerosene solution 1 per cent. kerosene solution	200 mg./kg. daily skin application 50 mg./kg. daily for 2 hours	All 5 dead after 14 applications. After 20 applications only 1 rat dead.
	DDT in kerosene, ether, Dimethylphthallate and Dibutylphthallate	Single doses of 3,000 mg./kg.	L.D. 50.
C) Smith & Stohlman, 1944	DDT in olive oil	150 mg./kg. orally	L.D. 50.
Woodard, Nelson & Calvery, 1944	DDT in corn oil	180 mg./kg.	L.D. 50.
	DDT	0·05 per cent. in diet	Signs of toxicity.
Cox, 1944 quoting Thienes	DDT	1 : 10,000 in food	White rats: convulsions after 2 months, died 3 days later.
Domenjoz, 1944	In gum arabic emulsions	0·5 gm./kilo. orally	L.D. 100.
	In olive oil	0·28 gm./kilo.	L.D. 100.
Draize and others, 1944	DDT in corn oil	200 mg./kg.	L.D. 50.
	DDT	400 p.p.m. for 50 weeks 800 p.p.m. for 50 weeks 500 p.p.m. in diet	Nervous symptoms. Half died in 2 weeks. Toxicity within a few weeks.

TABLE VIII, Sect. 2—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Rats—*continued.*	
(C) Cameron & Burgess, 1945	DDT in liquid paraffin	800 mg./kg. orally	L.D. 50.
Prill, Hartzell & Arthur, 1945	Rockland rat diet including DDT 0·2 per cent. and corn oil 2 per cent.	In diet	4 white rats, all died within 8–10 days.
Telford & Guthrie, 1945	DDT	Transmitted through goats' milk	Lethal to rats.
Läuger and others, 1946	DDT in oil	0·25 mg./kg. orally	3 out of 8 died, average duration of life 29 hours.
		0·50 mg./kg. and upwards orally	L.D.100.
Philips & Gilman, 1946	10 per cent. solution in peanut oil	200 mg./kg, orally	6/10 mortality.
		250 mg./kg. orally	22/25 mortality.
		300 mg./kg. orally	99/110 mortality.
		400 mg./kg. orally	10/10 mortality.
		500 mg./kg. orally	10/10 mortality.
(D) Woodard, Nelson & Calvery, 1944	Corn oil	Intra-muscular, intra-peritoneal, sub-cutaneous	Doses higher than for oral (180 mg./kg.) but action irregular.
Cameron & Burgess, 1945	Liquid paraffin	1,500 mg./kg. sub-cutaneous	L.D. 50.
Philips & Gilman, 1946	10 per cent. solution in peanut oil	Intravenous—	
		30 mg./kg.	1/10 mortality.
		40 mg./kg.	4/20 mortality.
		50 mg./kg.	21/33 mortality.
		60 mg./kg	12/14 mortality.
		Rabbits.	
(A) Cameron & Burgess, 1945	15 gm. in 30 cc. acetone	Conc. of 1 : 1,000 by air driven mechanical sprayer in cu. metre chamber exposed for 2 hours on 11 occasions	2 dead at 8 and 9 exposures.
(B) Draize, Nelson & Calvery, 1944	In corn oil	940 mg./kg. applied to skin	L.D. 50.
	In DMP*	2,820 mg./kg.	Ditto
	In DBP†	2,350 mg./kg.	Ditto
	In DMP* In DBP†	150 mg./kg./day for 30 days	Caused death.

*DMP=Dimethylphthallate. †DBP=Dibutylphthallate.

TABLE VIII, Sect. 2—*continued*.

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
Rabbits—*continued*.			
B) Draize and others, 1944	30 per cent. solution of DMP.*	In doses of 0·5, 1·0, 2·0 and 4·0 ml./kg. daily for 13 weeks applied to skin	Deaths at all levels.
Cameron & Burgess, 1945	In ether, kerosene, DMP* and DBP†	300 mg./kg.	L.D. 50.
	10 per cent. kerosene solution	Daily application of 100 mg./kg.	All 5 dead after 6 applications.
	1 per cent. kerosene solution	Daily application for 2 hours of 50 mg./kg.	None survived more than 16 applications.
	1 per cent. ethyl alcohol solution	10 mg./kg. for 2 hours daily	2/5 dead at 9 and 12 applications.
	1 per cent. acetone solution	Ditto	1/5 dead at 7 applications.
	1 per cent. ether solution	Ditto	1/5 dead at 13 applications.
Taylor, 1945	5 per cent. pure DDT in acetone	Applied to head, ears and feet	2-months-old rabbits. After 2 hours all severely affected, worst *in extremis* being killed and 1 died 2 days later, remaining 2 recovered. 8-months-old rabbits. 2 destroyed *in extremis* morning after experiment. 3rd killed 2nd day. 4th killed for P.M.
(C) Lindquist, Knipling, Jones & Madden 1944	DDT	537 mg./kg. orally	1 rabbit died.
Woodard, Nelson & Calvery, 1944	In corn oil	>400 mg/kg. orally	L.D. 50.
Domenjoz, 1944	Emulsion of gum arabic	0·275 gm./kilo. orally	L.D.
Smith & Stohlman, 1944	Olive oil	Stomach: 159 mg./kg. 200 mg./kg. 300 mg./kg.	 L.D. 33 per cent. L.D. 25 per cent. L.D. 50 per cent.
	Ditto	Chronic toxicity: 50 mg./kg. No. of doses 23 25 23 18 18 15	Total fatal dose gm./kg. 1·15 1·25 1·15 ·90 ·90 ·75 Average no. doses 20. Average total fatal dose 1·01 gm./kg.

*DMP=Dimethylphthallate. †DBP=Dibutylphthallate.

TABLE VIII, Sect. 2—*continued.*

	Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Rabbits—*continued.*		
(C)	Cameron & Burgess, 1945	Liquid paraffin 5 per cent. suspension in paraffin and tragacanth	Stomach: 300 mg./kg. repeated doses 50 mg./kg. by stomach tube. Total dosage 1·5 gm./kg.	L.D .50. 10 rabbits, 4 died at 8, 10, 11 and 20 doses.
(D)	Woodard, Nelson & Calvery, 1944	In corn oil	Intra-muscular, intra-peritoneal, subcutaneous	Doses higher than for oral >(400 mg./kg.) but action irregular.
	Cameron & Burgess, 1945	Liquid paraffin	Subcutaneous: 250 mg./kg.	L.D. 50.
	Philips & Gilman, 1946	DDT emulsion	Intravenous: 50 mg./kg. 75 mg./kg.	 L.D. 50. L.D. 100.
		Cats.		
(B)	Stage, 1946	5 per cent. emulsions 10 per cent. dusts	Dipping Dusting	Death. Death.
(C)	Smith & Stohlman, 1944	Olive oil	Stomach: 300 mg./kg.	L.D. 50.
(D)	Philips & Gilman, 1946	Peanut oil emulsion	Intravenous: 40 mg./kg. 50 mg./kg. 60 mg./kg. 75 mg./kg.	Mortality: 4 out of 4. 12 out of 16. 13 out of 17. 3 out of 3.
		Dogs.		
(B)	Hill & Robinson, 1945	5 per cent. kerosene	Sprayed once	Died 2 months later from exhaustion.
(D)	Philips & Gilman, 1946	DDT emulsion	75 mg./kg. inoculated	L.D. 100.
		Monkeys.		
(C)	Hill & Robinson, 1945	5 per cent. in kerosene	30 cc.	Died in 1 hour (2 baboons).
	Hill, 1946	5 per cent. in kerosene	27 cc. 30 cc.	L.D.
(D)	Philips & Gilman, 1946	As emulsion	Intravenous: 50 mg./kg. 60 mg./kg. 75 mg./kg.	Mortality: 1 out of 2 1 out of 2. 2 out of 2.

TABLE VIII, Sect. 2—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
		Man.	
Case, 1945	DDT distemper: Water 39 per cent., oil-modified phenolic resin 10 per cent., silicious matter, china clay, yellow ochre, Titanium dioxide 49 per cent. DDT 2 per cent., very small quantities of chlorinated phenol and subs. of ammonia	Two men, two periods each of 48 hours with intervals of 48 hours between. Both subjects wore short trunks only and sat so that large areas, chiefly back, were always in contact with the oily walls	Variety of unusual symptoms and physiological findings. Complete recovery after experiment.
Mackerras & West, 1946	DDT powder	Mixed in pastry in mistake for baking powder	Native cook boy in New Guinea made a tart eaten by some 25 men. All suffered from giddiness and weakness, beginning from 1 to 2½ hours after meal. Four vomited and two were taken to field ambulance; all recovered within 48 hours.
	DDT 5 per cent. solution in light diesel oil	Careless handling by malaria control man allowed right hand to become covered with solution on six days out of seven, probably for weeks	Finally developed numbness, muscular weakness and some swelling of right hand and suffered from severe deep-seated headache. About 2 weeks after onset of symptoms began to vomit at night and ran temperature of 101°F. No indication of liver involvement. Reported sick and went off duty. Symptoms disappeared in 4 days but 14 days before muscular power returned to normal.
	DDT powder	Dropped measure into tin and cloud of powder flew up, some of it getting into his eyes	Intense pain for 4 days requiring repeated injections of morphine and castor oil drops; blind for 2 weeks and severe headache for same period. Recovery complete.
Wigglesworth, 1945	DDT in acetone 25 gm. DDT in acetone with inert dust	Laboratory worker deliberately swabbed back of hand and prolonged contact of hands with DDT acetone; also kneaded	Some days later suffered from heaviness and aching in limbs with weakness in legs. Sleep almost impossible and acute mental anxiety.

TABLE VIII, Sect. 2—*continued.*

Author	DDT: Type and Formula	Dosage and Method of Application	Reason for Application and Remarks
	Man.—*continued.*		
Wigglesworth, 1945 —*cont.*		DDT/acetone dust with hands for some minutes	Took to bed and muscular tremors occurred through whole body. Recovery slow, incomplete at end of a year.
(C) Hill & Robinson, 1945	5 per cent. in kerosene	Drunk in error 1 oz. of 5 per cent. solution, dose approx. 105 mg./kg.	African child of 19 months; vomiting and convulsions followed by death in a few hours.
Daily Herald, 13/ii/46	DDT / kerosene fly spray	Suicide. Subject drank large quantity	50-year-old man who found his 79-year-old father accidentally killed by fumes from a stove, he then drank the DDT solution.
Biden-Steele & Stuckey, 1946 See also Daily Telegraph, 13/iv/46	20 per cent. in methylcyclohexanone	Suicide by swallowing mushroom fly emulsion. Total quantity taken found in stomach was 34 gms. DDT and 2·4 oz. methylcyclohexanone. This corresponds to 500 mg./kg. DDT.	Suggested that solvent assisted the toxic action of the DDT.
Neal, 1946	6 per cent. in kerosene	Drunk by a woman	Dead within 3 hours.
Evening Standard, 27/viii/46	DDT. Highly concentrated solution	Accidental inhalation of concentrated fumes in a summer house	Subject was trying to get rid of spiders in his summer house. Death resulted.
U. Chit Thoung, 1946	DDT in rice	One of two samples of rice taken in meal gave 16 per cent. of DDT the other one was free	Frontier Force constabulary at Wawang near Bhamo on July 29th, 1946, suffered from vomiting and purging about 1 hour after evening meal. Some white particles found in rice. 27 men who had vomited several times also complained of giddiness. The following morning the District Health Officer went to camp and found 72 men suffering from food poisoning. Typical symptoms: low pulse, 40–50/minute, giddiness when getting up and dilated pupils.

The foregoing data may be further analysed :—

The small experimental laboratory animals are dealt with in Tables IX to XII. In Table IX the salient facts from Section 2 of Table VIII are arranged according to the principal solvents or carriers used. Table X is an analysis of toxic cases in relation to the type of exposure, solvent or carrier and animal, while Table XI is a summary of the toxic doses recorded with the principal solvents, classified under method of exposure.

TABLE IX.

Analysis of Toxic Data : Solvent/Carrier-Dosage-Animal.

Solvent or Carrier, Dosage and Method of Application	Guinea Pig	Mouse	Rat	Rabbit	Cat	Dog	Monkey
Acetone. *Aerosol.* 1 : 1,000 thick mist	X	X	X	X			
Skin Exposure. 1 per cent., 10 mg./kg. daily for 2 hours ... ... (G.P. dead at 7 and 9 applications, 2/5.) (Rabbit dead at 7 applications, 1/5.)	X			X			
5 per cent. to head, ears and feet				X			
Cyclohexanone. *Aerosol.* Freon : daily exposure 0·183 mg./litre air every 15 minutes for 45 minutes three times/day, 5 days/week/4 weeks		X					
Sesame Oil. *Aerosol.* Freon : 12·44 mg./litre air for 45 minutes		X					
Dimethylphthallate. *Skin Exposure.* 150 mg./kg./day/30 days	X		X	X			
Doses of 0·5, 1·0, 2·0, 4·0 mg./kg. daily for 13 weeks, deaths at all levels	X		X	X			
Single doses : 3,000 mg./kg. L.D. 50			X				
2,820 mg./kg. L.D. 50				X			
300 mg./kg. L.D. 50				X			

TABLE IX—*continued.*

Solvent or Carrier, Dosage and Method of Application	Guinea Pig	Mouse	Rat	Rabbit	Cat	Dog	Monkey
Dibutylphthallate. *Aerosol.* 54·4 mg./litre air		X					
Skin Exposure. 150 mg./kg./day for 30 days ...				X			
Single doses: 3,000 mg./kg. L.D. 50			X				
2,350 mg./kg. L.D. 50				X			
300 mg./kg. L.D. 50				X			
Kerosene. *Skin Exposure.* 10 per cent. solution 200 mg./kg., 12 applications	X						
10 per cent. solution 200 mg./kg., 14 applications			X				
10 per cent. solution 100 mg./kg., 6 applications				X			
1 per cent. solution 50 mg./kg./day for 2 hours— 16 applications				X			
18 applications	X						
Single doses: 3,000 mg./kg. L.D. 50			X				
1,000 mg./kg. L.D. 50	X						
300 mg./kg. L.D. 50				X			
5 per cent. spray						X	
Oral. 25–30 cc. 5 per cent. solution ...							X
Ether. *Skin Exposure.* 10 mg./kg. 2 hours daily, 1 per cent. (Guineapig 2/5 dead at 7 and 8 applications.) (Rabbit 1/5 dead at 13 applications.)	X			X			
Single doses: 3,000 mg./kg. L.D. 50			X				
1,000 mg./kg. L.D. 50	X						
300 mg./kg. L.D. 50				X			

TABLE IX—*continued.*

Solvent or Carrier, Dosage and Method of Application	Guinea Pig	Mouse	Rat	Rabbit	Cat	Dog	Monkey
Corn Oil.							
Skin Exposure. 940 mg./kg. . . . L.D. 50 ...				X			
Oral. 180 mg./kg. . . . L.D. 50 ...			X				
200 mg./kg. . . . L.D. 50 ...			X				
400 mg./kg. . . . L.D. 50 ...	X	X					
400+ mg./kg. . . L.D. 50 ...				X			
448 mg./kg. . . . L.D. 50 ...		X					
Inoculation. 400+ mg./kg. . . . L.D. 50 ... Intramuscular, intraperitoneal, subcutaneous.				X			
448+ mg./kg. . . . L.D. 50 ...		X					
480+ mg./kg. . . . L.D. 50 ...			X				
562 mg./kg. . . . . L.D. 50 ...	X						
Liquid Paraffin.							
Oral. 50 mg./kg. 8–20 doses				X			
400 mg./kg. . . . L.D. 50 ...	X						
800 mg./kg. . . . L.D. 50 ...			X				
300 mg./kg. . . . L.D. 50 (Stomach)				X			
Inoculation. 250 mg./kg. . . . L.D. 50 ...				X			
900 mg./kg. . . . L.D. 50 ...	X						
Subcutaneous, 1,500 mg./kg. L.D. 50			X				
Gum Arabic.							
Oral. 0·275 gm./kg. . . . L.D. 50 ...				X			
0·5 gm./kg. . . . L.D. 50 ...			X				
1·6 gm./kg. . . . L.D. 50 ...		X					
Olive Oil.							
Oral. 0·175 gm./kg. toxic		X					
0·28 gm./kg. toxic			X				
150 mg./kg. . . . L.D. 50 ...			X				

TABLE IX—*continued.*

Solvent or Carrier, Dosage and Method of Application	Guinea Pig	Mouse	Rat	Rabbit	Cat	Dog	Monkey
Olive Oil—*continued.* *Oral.—cont.*							
159 mg./kg. . . . L.D. 33 ...				X			
200 mg./kg. . . . L.D. 25 ...				X			
300 mg./kg. . . . L.D. 50 ...				X	X		
Chronic toxicity. 50 mg./kg. average number doses 20, Total average fatal dose 1·01 gm./kg.				X			
Peanut Oil. *Oral.* 10 per cent. solution. 200 mg./kg. 6/10 mortality			X				
10 per cent. solution. 250 mg./kg. 22/25 mortality			X				
10 per cent. solution. 300 mg./kg. 99/110 mortality			X				
10 per cent. solution. 400 mg./kg. 10/10 mortality			X				
10 per cent. solution. 500 mg./kg. 10/10 mortality			X				
Inoculation. Intravenous. 30 mg./kg. 1/10 mortality			X				
Intravenous. 40 mg./kg. 4/20 mortality			X		X4/4		
Intravenous. 50 mg./kg. 21/23 mortality			X		X12/16		
Intravenous. 60 mg./kg. 12/14 mortality			X		X13/17		
Intravenous. 75 mg./kg. ...					X3/3		
Ethyl Alcohol. *Skin Exposure.* 10 mg./kg. 2 hours daily. 2/5 at 9 and 12 applications				X			
Oil. *Oral.* 0·25 mg./kg. 3/8 average 29 hours			X				
0·50 mg./kg. L.D. 100			X				

TABLE IX—*continued.*

Solvent or Carrier, Dosage and Method of Application	Guinea Pig	Mouse	Rat	Rabbit	Cat	Dog	Monkey
DDT. *Oral.* 0·05 per cent. in diet		X					
Below 500 p.p.m. in diet		X					
1 : 10,000 diet			X				
500 p.p.m. in diet			X				
400 p.p.m. for 50 weeks			X				
800 p.p.m. for 50 weeks			X				
537 mg./kg.			X				
Miscellaneous. *Oral.* Rockland Rat Diet with Corn Oil 2 per cent. and DDT 0·2 per cent. L.D. 10 days			X				
Milk of Goat fed DDT			X				
Aerosol. Primary solvent and secondary oil solvent 3 per cent.			X				
Skin Exposure. 5 per cent. emulsion. Dipping . . . L.D. 50					X		
10 per cent. Dusts					X		
Inoculation. Intravenous. 50 mg./kg. L.D. 50				X			X1/2
Intravenous. 75 mg./kg. L.D. 100				X		X	X2/2

Also 0·05 per cent. in diet of young chicks and 0·5 gm. in capsules for pigeons were both toxic.

TABLE X.

Toxicity, Type of Exposure, Solvent/Carrier and Animal.

AEROSOLS.

Solvent or Carrier	Guinea Pig	Mouse	Rat	Rabbit	Cat	Dog	Monkey	Totals
Acetone	1	1	1	1	—	—	—	4
Cyclohexanone ...	—	1	—	—	—	—	—	1
Sesame Oil ...	—	1	—	—	—	—	—	1
DBP	—	1	—	—	—	—	—	1
Miscellaneous ...	—	—	1	—	—	—	—	1
Totals ...	1	4	2	1	—	—	—	8

TABLE X—*continued.*

SKIN EXPOSURE.

Solvent or Carrier	Guinea Pig	Mouse	Rat	Rabbit	Cat	Dog	Monkey	Totals
Acetone	1	—	—	2	—	—	—	3
DMP	2	—	3	4	—	—	—	9
DBP	—	—	1	3	—	—	—	4
Kerosene... ...	3	—	2	3	—	1	—	9
Ether	2	—	1	2	—	—	—	5
Corn Oil	—	—	—	1	—	—	—	1
Miscellaneous ...	—	—	—	1	2	—	—	3
Totals ...	8	—	7	16	2	1	—	34
ORAL.								
Kerosene... ...	—	—	—	—	—	—	2	2
Liquid Paraffin ...	1	—	1	2	—	—	—	4
Gum Arabic ...	—	1	1	1	—	—	—	3
Corn Oil	2	2	2	1	—	—	—	7
Olive Oil	—	1	2	4	1	—	—	8
Peanut Oil ...	—	—	5	—	—	—	—	5
Miscellaneous ...	—	—	4	—	—	—	—	4
DDT	—	2	5	—	—	—	—	7
Totals ...	3	6	20	8	1	—	2	40
INOCULATION.								
Liquid Paraffin ...	1	—	1	1	—	—	—	3
Corn Oil	1	1	1	1	—	—	—	4
Peanut Oil ...	—	—	4	—	4	—	—	8
Miscellaneous ...	—	—	—	2	—	1	2	5
Totals ...	2	1	6	4	4	1	2	20
Animals: totals	14	11	35	29	7	2	4	102

Total number of cases, 102 :

Acetone	7	Liquid Paraffin ...	7
Cyclohexanone ...	1	Gum Arabic ...	3
DMP	9	Corn Oil	12
DBP	5	Olive Oil	8
Sesame Oil ...	1	Peanut Oil	13
Kerosene	11	Miscellaneous ...	13
Ether	5	DDT	7

TABLE XI.

Analysis of Toxic Doses.

SKIN EXPOSURE. L.D. 50. The figures indicate mg./kg. body weight. Single Doses.

Animal	DMP (1)	DBP (2)	Kerosene	Ether	Corn Oil
Guinea Pig	—	—	1,000	1,000	—
Rat	3,000	3,000	3,000	3,000	—
Rabbit	2,820 300	2,350 300	— 300	— 300	940

(1) Dimethylphthallate. (2) Dibutylphthallate.

ORAL. L.D.50.

Animal	Corn Oil	Liquid Paraffin	Gum Arabic	Olive Oil	Peanut Oil
Guinea Pig	400–562	400	—	—	—
Mouse ...	400–448	—	1,600	175	—
Rat ...	180–200	800	500	150–280	200
Rabbit ...	400+	300	275	300	—

INOCULATION. L.D.50.

Animal	Corn Oil	Liquid Paraffin	Peanut Oil	5% Emulsion
Guinea Pig	562	900	—	—
Mouse	448+	—	—	—
Rat	180+	1,500	40–50	—
Rabbit	400+	250	—	50

The figures for aerosols are too indeterminate to be included in this table.

It is difficult to deduce any conclusive facts from the evidence so far published. Animal species differ considerably in their reaction to DDT, irrespective of route and administration. While it is probably true to say that the larger species show greater relative resistance, among the smaller laboratory animals the results are conflicting, not only between different species but, in some cases, among the same species. Thus the records from cats and rabbits are most contradictory; with the former, Smith & Stohlman (1944) and Philips & Gilman (1946) show markedly uneven results, and with rabbits the L.D.50 recorded by Draize, Nelson & Calvery (1944) show a much higher resistance (2,820 mg./kg. DMP, 2,350 mg./kg. DBP and 940 mg./kg. corn oil) than those of Woodard, Nelson & Calvery (1944) (>400 mg./kg. corn oil) or Cameron & Burgess (1945) (300 mg./kg. ether, kerosene, DMP & DBP). Similarly it is difficult to form conclusions as to the significance of the principal solvents used. The figures speak for themselves and further comment is unnecessary.

Many factors may account for such discrepancies; the breed of animal, the condition of the animals at the time of the experiment, local conditions of climate and environment, etc. Unfortunately, much of the literature lacks essential data in varying degree and it is only possible to take a general view of the work published.

When we come to consider the larger animals the position is somewhat different. The L.D.50 of goats, sheep, pigs, horses and cattle has not been determined and the data on dosage is much less precise as most of the work has been done on a field scale where accurate measurement is impossible. There are, therefore, no records of toxic results known to the writers, apart from temporary effects with subsequent recovery. Telford & Guthrie (1946), Cox (1944), Neal & von Oettingen (1946) and Stage (1946).

Table XII shows the maximum non-toxic doses to the principal experimental laboratory animals. Comparison of the three factors, animal, method and solvent, is here again difficult but it certainly indicates that the minimum lethal dosage is very much higher for animals than it is for insects.

TABLE XII.

Maximum Non-toxic Doses to Principal Experimental Animals.

Nature of exposure	Guinea Pig	Mouse	Rat	Rabbit	Dog
AEROSOLS.					
1 : 3,000 thick mist		X	X	X	
12·22 mg./litre air for 45 mins.		X			X
12·44 „ „ „	X		X		
32·9 „ „ „	X		X		X
54·4 „			X		
Heavy mist for 48 mins. daily for 4 weeks				X	
Repeated single doses of 32 mg./litre air					X
Continuous-phase aerosol by electrical volatilisation approx. 3·4 mg./cu.ft. for 28 days. Total output 21,000 mg.			X	X	
SKIN EXPOSURE.					
150 mg./kg. over 14 days	X		X		
300 mg./kg. over 12 days	X		X		
75 cc. in 15 doses				X	
5 per cent. in pyrophyllite	X				X
5 per cent. aqueous emulsion	X				X
5 per cent. vaseline	X				
10 per cent. pyrophyllite					X

TABLE XII—*continued.*

Nature of exposure	Guinea Pig	Mouse	Rat	Rabbit	Dog
ORAL.					
100 mg./kg.			X		X
500 mg./kg.		X			
537 mg./kg.				X	
200–400 p.p.m. in diets			X		
1,200 mg. over 6 days			X		
50 mg./kg./30 doses, total 1,590 mg.			X		
2,640 mg. over 17 days				X	
50 mg./kg./day/300 days, total 15,000 mg.					X
INOCULATION.					
Intramuscular: 15 mg./kg. over 5 days		X			
Intravenous: 25 mg./kg.				X	
Intravenous: 60 mg./kg.					X

Wild Life.

Very little evidence is available on the toxicity of DDT to wild life. Such data as is known to the writers, principally concerning aquatic life, is given in Table VIII. From this and from the general entomological literature and the writers' and others' experience, it would seem that the margin between toxic doses for mosquito and other aquatic insect larvae and for crustacea, batrachians and fish is appreciably narrower than that between animals and insects. Much work is necessary before definite dosages for different types of water, climate and more specific environment can be postulated with any certainty. A typical example of the possibilities resulting from injudicious use of DDT on a large scale is given by Stage (1946) in which he cites the case of the destruction of an estimated 100,000 small fish owing to the accidental release of an overdose of aerial spraying against mosquito larvae.

Conclusions from the Literature on the Toxicity of DDT to Animals.

In considering the available evidence from every angle a clear conclusion that DDT is non-toxic to animals, and in particular to the larger domestic animals, in normal insecticidal doses may be stated with every confidence. It may be of interest, moreover, in this connection, to quote some definite statements from the "Minutes of a Conference on Toxicity of DDT to Man and other Mammals and the Hazards involved in the Agricultural Use of DDT" recently held by the Insect Control Committee, National Research Council, Washington. Decker (1946) states "Toxicologists agree that the acute and sub-acute toxicity of DDT to warm-blooded animals is no greater, and some say very much less, than that of several materials now in common use as insecticides. The U.S. Food and Drug Administration has for all practical purposes acknowledged this fact in establishing its tentative administrative tolerance of 7 p.p.m. (about 0·05 grain per pound) for DDT on apples and pears. . . . In the case of animals there is considerable biological data, all of which tend to show that cattle, swine, sheep, goats, chickens and laboratory animals can, over long periods of time, ingest daily amounts of DDT far in excess of what might be expected as a residue on plants without showing any ill effects. The writer has no knowledge of even a single authentic recorded case of animal poisoning as a result of their feeding on DDT-treated plants."

A note of warning is, however, sounded by Draize (1946) at the same conference ; " It is now established that animals eating forage crops sprayed with DDT transmit this poison in the milk and also store significant quantities in the meat, particularly in the fat portion. . . . The animal experiments permit the following conclusions :—

1. The hazards from contact of DDT powders with the skin are insignificant.
2. There appears to be little hazard from the inhalation of aerosols, mists, sprays or dusts.
3. DDT may be absorbed from solutions following skin contact to produce fatal poisoning.
4. The wide individual variation in susceptibility to poisoning, regardless of the route of administration, makes it difficult to establish safe tolerance limits to cover all individuals.
5. The tentative tolerance of 7 p.p.m. which the Food & Drug Administration has set for apples and pears may have to be revised if man is to be exposed to DDT from other sources, *e.g.* meats, dairy products, etc."

The two statements are not contradictory and while it is clear that little harm is likely to come to domestic animals as the result of normal insecticidal activities employing DDT it is equally clear that the implications in Draize's statement require further investigation.

The Toxicity of DDT to Man.

The accurate assessment of lethal dosage, where man is concerned, is impossible at the present time as subjects have a scientifically regrettable reluctance to pursue investigations to a logical conclusion. The known evidence is, to some extent, confusing but on balance it is fairly clear that only gross carelessness can result in serious harm or death from DDT. The cases in which toxic results have occurred fall into two classes : those in which toxic effects have resulted but with eventual recovery and those which have resulted in death.

In the former category, Case (1945) and Wigglesworth (1945) furnish the only evidence from the laboratory, while Mackerras & West (1946) record three cases from the New Guinea war zone, and Chit Thoung (1946) records the poisoning of 72 men, 27 relatively acutely, of a Frontier Force Constabulary at Wawang near Bhamo from eating rice contaminated with 16 per cent. DDT in powder form. (See Table VIII, Sect. 1, pp. 53–54, under Man.)

Case (1945) has described an investigation in which he and his technician spent 48 hours in contact with an oily solution of DDT. The investigation was conducted in a steel chamber, octagonal in shape, 6 ft. across and 6 ft. high. They alternately sat and slept on a narrow seat running round the inside and in doing so a large skin area necessarily made contact with the walls. After a first sojourn of 48 hours the walls were painted with a DDT distemper (see Table VIII, Sect. 2) on top of which was placed a covering film of lubricating oil. Two days after this application to the walls they spent a second 48 hours in the chamber as a result of which the following toxic effects were noted :—

An increase in erythrocyte destruction, a decrease in the mean corpuscular haemoglobin, an increase in reticulocytes, a slight fall in the haemodynamic index, a diminution of polymorphonuclear leucocytes, an increase in lymphocytes accompanied by the appearance of immature white cells, a fall in systolic blood pressure, the presence of indoxyl sulphate in the urine, heaviness and aching of the limbs, diminution of some reflexes, unilateral slight impairment of hearing, transient yellow vision (one subject), muscular fibrillation (one subject), peripheral patchy anaesthesia (one subject) weakness of the legs, a fine tremor (one subject), changes in the mental state with extreme irritability and violent joint pains. One of the men felt so ill that he had to take to his bed for a day. Case suggests that this experiment only shows DDT to be toxic under certain conditions and that there should be no condemnation of the use of DDT but that proper precautions should be taken, especially against oily contamination.

Wigglesworth (1945) described a laboratory worker who on at least two occasions deliberately allowed an acetone DDT solution to evaporate on the back of his hands. He also kneaded a DDT mixture with his hands for some minutes. Some days later he suffered from heaviness and aching in his limbs with weakness in his legs. Sleep became almost impossible and a state of acute mental anxiety developed. He had to take to his bed, during which time muscular tremors occurred throughout his whole body. When he was told that his was undoubtedly a case of DDT poisoning, he immediately began to improve. His recovery was slow and was not complete at the end of a year. Wigglesworth suggests that the man's exposure was far in excess of anything likely to occur in practice.

Mackerras & West (1946) cite three cases of toxic effects from exposure to DDT, one from eating pastry in which DDT powder had been mixed by mistake (25 men), one from careless handling by a malaria control man mixing solution and one from accidental exposure of the eyes to DDT powder. (See Table VIII, Sect. 2.)

Cases of Death to Man.

There are five cases known to the writers (Table VIII, Sect. 2). The first recorded one is the now well-known case of the African child written up by Hill & Robinson (1945). The next was reported in the " Daily Herald " of 13/ii/46, in which a suicide drank a large quantity of DDT/kerosene fly spray, the third was reported in the " Daily Telegraph " of 13/iv/46, presumably another suicide, in which the victim swallowed a quantity of mushroom-fly emulsion ; this was written up by Biden-Steele & Stuckey (1946). They record that the victim died some time within an hour after drinking the emulsion. The stomach held about 6 oz. of the emulsion which contained 20 per cent. DDT dissolved in 40 per cent. methylcyclohexanone. They calculated that the total quantity of DDT and methylcyclohexanone taken was at least 34 grams and 2·4 oz. by weight respectively, these being the amounts found in the stomach. This they consider corresponds to a concentration of 500 mg./kg. body weight. They state that methylcyclohexanone is not a recognised poison and that they have been unable to find a case of poisoning due to its administration. They consider that death was primarily due to DDT but suggest that the solvent assisted the toxic action.

Neal (1946) mentions the case of a woman who drank 6 per cent. DDT in kerosene and died within three hours ; no further details are available. Finally, in the " Evening Standard " of 27/viii/46 the accidental death of a man in Collingwood, N.J., U.S.A., was reported ; apparently he was overcome by the fumes from a highly concentrated solution of DDT with which he was attempting to rid his summer house of spiders.

Exposure of Man to DDT with no Harmful Results.

The evidence for the other side of the argument is much more impressive. No less than thirteen cases, varying from 62 men to one child as subjects, have been recorded of DDT contamination of greater or lesser extent, without harmful results. The data are recorded in Table VIII, Sect. 1, and need not be repeated here, but it is of interest to consider the salient facts that arise from a study of these cases.

We may leave the cases of Wasicky & Unti (1944) and Neal (1946) out of consideration as in the former the exposure to DDT does not seem to have been very great and in the latter, the child's stomach was washed out promptly thus negativing any results. Of the remainder, two cases, Anon., 1945a (see p. 42) and Cameron & Burgess (1945) (p. 42), record extensive trials on 62, and 12, 40 and 6 men respectively. In the former case a large selection of formulae and solvents was tried and exposure was very much greater than any recorded with toxic results.

The writers' contribution is significant primarily from the fact that the subjects investigated were working throughout under operational conditions, and that full data of environment and contamination are given, the latter being greatly in excess of anything described by Wigglesworth or Case.

Neal and others (1944), Neal, von Oettingen, Dunn & Sharpless (1945), Neal, Sweeney, Spicer & von Oettingen (1946), C.D.E.S., Porton (unpublished) and Willoughby & Sheehan (unpublished) provide strong laboratory evidence which is in direct contradiction to that of Case & Wigglesworth, and MacCormack (1945), Lazar (1946) and Neal, Sweeney, Spicer and von Oettingen (1946) furnish evidence of the ingestion of DDT by man, the quantities being 1,500 mg., unknown, and 770 mg. respectively.

Finally, Fennah (1945) has exposed himself to the inhalation (100 mg. pure sifted DDT) and ingestion (water dusted at more than 300 mg./sq. ft.) for approximately 12 months and later to the further ingestion (spraying of all food taken with 3 per cent. emulsion) and to the exposure of hands and arms to 5 per cent. emulsion, giving a dosage of over 200 mg./sq. ft. (See Table VIII, Sect. 1, p. 44, under Man.)

Discussion.

A review of the knowledge that is available to date leads to no clear-cut picture, but the evidence of the toxicity of DDT to man is far from convincing. Although there is no case on record, it must be concluded that DDT by itself would cause toxic effects if it were taken by mouth in sufficient quantities. If a man dies as a result of drinking a DDT emulsion or solution it would not seem possible to exclude the solvent itself as the toxic agent. Uncertainties of this kind are inevitable where more than one chemically pure substance is administered. Thus, in considering the case reported by Hill & Robinson (1945), it should be remembered that kerosene, on a number of occasions, has been shown to be capable of causing death (Balaban, 1946). It is not without interest in this connection to consider the list of substances that have been used as solvents or carriers of DDT in liquid or aerosol form without harm to animals. Kerosene, alcohol, benzene, acetone, benzyl benzoate, toluene, turpentine, vaseline, ammonia, hexamethylenetetramine, cyclohexanone, dimethylphthallate, dibutylphthallate, sesame oil, castor oil, corn oil, olive oil, peanut (arachis) oil, pine oil, liquid paraffin, gum tragacanth and a number of commercial solvents and detergents have been used. Calvery & Neal (1946) state that of the hydrocarbon solvents, kerosene, fuel oil, tetrahydronaphthalene, xylene, heavy solvent, PD544-C, Velsicol AR-50, K-327, S/V Culicide oil, Velsicol NR-70, APS-202, Hi-flash solvent, and Solvesso No. 3, some have not been studied, some are being studied and some have been thoroughly investigated. All introduce a dermatitis hazard as well as possible toxicity from inhalation and skin absorption. Non-hydrocarbon solvents cyclohexanone, o-dichlorobenzene and isophrone, all of which have been investigated toxicologically, are considered safe if the recommended precautions are followed. It is stated that there is no hazard from ingestion and little from skin absorption and inhalation with emulsifiers (surface-active agents) of which a list of proprietary substances is given.

Both Wigglesworth and Case have shown that the presence of a solvent can indeed result in absorption and far reaching effects although it is surprising that they were able to demonstrate toxic effects, for it is clear that their subjects received only relatively light exposure. The correlation of their results with those of the workers mentioned above and of our own is not easy for the contrast is so striking, and it is possible that the explanation may be found to be due to intolerance or idiosyncrasy. In considering Wigglesworth's case, it is of interest to note the results obtained by C.D.R.E., India, extracts from which were published by Dangerfield (1946). The following is quoted from the original, " A special series of experiments was carried out with acetone solution simulating the circumstances of a case described by Wigglesworth though the exposures used here were probably considerably more severe. . . . The conditions of exposure described by Wigglesworth have been reproduced as far as possible. . . . In all experiments there were no toxic symptoms or signs attributable to DDT, and no abnormality was found in any specimen of urine examined." The writers feel that Dangerfield's view that " it seems possible that the patient was suffering from an anxiety neurosis " is a likely explanation.

For Case's work the environment chosen was a steel chamber, in which Case and his assistant sat and slept on a narrow steel seat which was placed against the inside walls. They remained for two periods of 48 hours in this chamber, the small size and uncomfortable furniture of which making it seem possible that superimposed on the real discomfort of their limbs was an anxious anticipation of their fate as a result of their contact with DDT, an anxiety that induced so many symptoms. Case states that both he and his technician, subject A.1, complained of smarting eyes and a sore throat, tiredness and pain in the limbs and a general feeling of illness which was so marked that the latter had to take to his bed. Case goes on to thank A.1 for his careful and conscientious examination of material. If this material was his own, the question arises as to the extent of the psychological bias involved in such a procedure. Nor would it seem possible for a man as sick as was described to make accurate observations, or indeed observations of any kind, for Case goes on to say, both for himself and his technician, that " not only was there a state of extreme irritability present but also both subjects had a great distaste for work of any sort and a feeling of mental incompetence in tackling the simplest mental task."

Conclusions.

In presenting our conclusions we think it not inappropriate to quote from eminent authority on both sides of the Atlantic.

Buxton (1945a) in a special note to the " Tropical Diseases Bulletin " said " DDT is of such immense importance that the question of its toxicity to man has been carefully looked into in the U.S.A. and Britain. I believe that men engaged in manufacture and those who have sprayed it for long periods have been carefully examined, and no evidence of toxic action of any kind found in them. There seems to be no evidence that DDT, used as an insecticide, is harmful to man or domestic animals."

Cameron & Burgess (1945) stated " It is apparent from this summary of American investigations and the results of our own experiments that animal tests, though indicating the toxic features of DDT, make it clear there is a wide margin of safety in its use as an insecticide. We have always maintained that provided a maximum concentration of 0·5 per cent.* be insisted upon for sprays there is no reason to anticipate any danger to man. . . . Even with long-continued exposure to such sprays it is difficult to see how ill effects could be incurred."

Neal (1946) at the Conference on Toxicity of DDT to Man and other Mammals and the Hazards incurred in the Agricultural use of DDT (May 1946) expresses his opinion as follows : " It may be pointed out that any solvent suitable for this purpose

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* Percentages up to 5 per cent. in liquids and 10 per cent. in powders are now commonly used with perfect safety.

(aerosols, smokes and power sprays) is liable to cause irritation of the skin, due to the same physico-chemical properties which render it valuable as a DDT solvent. For the same reason they cause irritation of the mucous membranes, and in sufficient concentrations may cause systemic effects such as nausea, vomiting, fatigue, headache, and other nervous system manifestations. . . . Since the introduction of the use of DDT by the armed forces, the Army, the Navy, the Bureau of Entomology and Plant Quarantine, U.S. Department of Agriculture, the Food and Drug Administration, the Office of Scientific Research and Development, the U.S. Public Health Service and other groups have brought to the attention of this laboratory (Nat. Inst. Health, Bethesda, Maryland), all suspected cases of DDT poisoning in humans. This laboratory has traced each case brought to its attention, and in some instances has hospitalised the individuals for intensive study. To date no cases have been discovered in which the signs and symptoms were considered to be due to DDT *per se*. In several cases the solvent for DDT was found to cause irritation of the eyes and upper respiratory tract, as well as such symptoms as headache, nausea, tingling sensations of the lips and face, nervousness or jittery feeling, listlessness and loss of equilibrium. In view of the extensive use of DDT as an insecticide, well over a million humans have been exposed to DDT. It is obvious that some of these will develop diseases, the true character of which are not readily diagnosed, and therefore easily credited to exposure to DDT. In summary it may be stated that DDT is definitely less toxic than Paris Green or sodium fluoride. Nevertheless, DDT has toxic properties which require certain precautions in its use."

There is little to add. In the light of our own experience and the data presented in this paper we consider that DDT when used as an insecticide, with reasonable intelligence and the precautions normal to the use of modern insecticides, is harmless to man and animals. It is, neverthless, possible by unskilled formulation, by the use of unsuitable solvents and by misuse in its application, to incur risk to man and animals. The possibilities of cumulative effects from the storage of DDT in the milk and tissues of sheep and cattle require further investigation.

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FURTHER NOTES ON THE FOOD-PLANTS OF NIGERIAN INSECTS, VI.*

By F. D. Golding, M.A., F.R.E.S.

d 2988 E.M.I

The present paper, which is the last of the series, contains additional data obtained between November 1939 and December 1945.

In 1939, *Helopeltis bergrothi*, Reut., was found feeding on cacao pods and shoots at Owena by the writer; it was evident that this species was distinct from the cotton *Helopeltis*, which had been identified as *bergrothi* by the Imperial Institute of Entomology in 1924. The species attacking cotton is *H. sanguineus*, Popp., and this name should be substituted for *bergrothi* in the three papers on food-plants published in 1935, 1937 and 1940.

The writer wishes to acknowledge his indebtedness to the Imperial Institute of Entomology and the specialists of the Department of Entomology in the British Museum for identifying the insects mentioned.

Name	Food-plants	Part attacked	Month of appearance	Locality and remarks
	LEPIDOPTERA			
NYMPHALIDAE—				
Acraea zetes, L.	*Adenia lobata*	Leaves	Feb.	Ibadan, larvae
A. terpsichore, L.	*Corchorus olitorius*	,,	July, Aug.	,, ,,
Catuna crithea, Drury	*Malacantha alnifolia*	,,	June	,, ,,
LYCAENIDAE—				
Lampides boeticus, L.	*Vigna unguiculata*	Pods	Feb.	Jos, ,,
HESPERIIDAE—				
Pteiroteinon laufella, Hew.	*Elaeis guineënsis*	Leaves	Dec.	Ibadan, ,,
ARCTIIDAE—				
Diacrisia maculosa, Cram.	Onions	,,	July	,, ,,
NOCTUIDAE—				
Calpe triobliqua, Saalm.	*Cissampelos owariensis*	,,	April	,, ,,
Dermaleipa metaphaea, Hmps.	*Mangifera indica*	Fruits	June	,, adults
Earias biplaga, Wlk.	*Abutilon mauritianum*	,,	March	,, larvae
E. insulana, Boisd.	,, ,,	,,	,,	,, ,,
Phytometra acuta, Wlk.	*Mucuna aterrima*	Leaves	April	,, ,,
Phytometra ni, Hb.	Cabbage	,,	March	Maiduguri, ,,
Plusia orichalcea, F.	Kohl rabi	,,	Sept.	Ibadan ,,
Prodenia litura, F.	*Solanum melongena*	,,	April	,, ,,
	Capsicum frutescens	Pods	Feb.	,, ,,
	Basella alba	Leaves	Sept.	,, ,,
	Cabbage	,,	Aug.	,, ,,
	Tomato	,,	Oct., Nov.	,, ,,
Xanthodes graellsii, Feisth.	*Hibiscus esculentus*	,,	April	,, ,,
Heliothis armigera, Hb.	Tomato	,,	Oct.	,, ,,

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*The previous five parts have appeared in the *Bulletin of Entomological Research*: Part 1 in **18**, Part 2 in **22**, Part 3 in **26**, Part 4 in **28**, and Part 5 in **31**.

Name	Food-plants	Part attacked	Month of appearance	Locality and remarks
	LEPIDOPTE	RA—*continued.*		
LYMANTRIIDAE—				
Naroma signifera, Wlk.	*Ficus thonningii* ...	Leaves ...	Sept. ...	Ibadan, larvae
Olapa tavetensis, Holl.	*Ochroma lagopus* ...	,, ...	Oct. ...	,, ,,
Dasychira remota, Druce	*Antiaris africana* ...	,, ...	April ...	,, ,,
SPHINGIDAE—				
Acherontia atropos, L.	*Solanum melongena*	,, ...	July ...	,, ,,
Deilephila nerii, L. ...	*Cinchona* sp. ...	,, ...	March ...	Bamenda, larvae
Hippotion celerio, L.	*Boerhaavia diffusa*	,, ...	Dec. ...	Ibadan ,,
Polyptychus contraria, Wlk.	*Spathodea campanulata*	,, ...	April ...	,, ,,
NOTODONTIDAE—				
Anaphe reticulata, Wlk.	*Afrormosia laxiflora*	,, ...	July ...	Gombe, ,,
	Terminalia avicennioides	,, ...	,, ...	,, ,,
Epanaphe moloneyi, Druce	*Isoberlinia doka* ...	,, ...	June, July	Zaria, ,,
E. vuilleti, de Joann.	*Tamarindus indica*	,, ...	July ...	,, ,,
GEOMETRIDAE—				
Neocleora tulbaghata, Feld.	Carrot	,, ...	Sept. ...	Ibadan, ,,
SATURNIIDAE—				
Epiphora bauhiniae, Guér.	*Ziziphus spina-christi*	,, ...	,, ...	Gombe, ,,
	Z. mauritiana ...	,, ...	,, ...	,, ,,
LASIOCAMPIDAE—				
Gastroplakaeis rufescens, Aur.	*Isoberlinia doka* ...	,, ...	Aug. ...	Zaria, ,,
Gonometa titan, Holl.	*Cassia nodosa* ...	,,	?	Umuahia, larvae
Gonombombyx angulata, Aur.	*Cola caricifolia* ...	,, ...	May ...	Ibadan, larvae
Pachypasa honrathi, Dew.	*Alchornea cordifolia*	,, ...	July ...	,, ,,
PYRALIDAE—				
Agathodes ostentalis, Hb.	*Erythrina* sp. ...	,, ...	May ...	,, ,,
Corcyra cephalonica, Staint.	*Elaeis guineënsis* ...	Nuts ...	April ...	,, ,,
Crocidolomia binotalis, Zell.	Turnip	Leaves ...	Aug. ...	,, ,,
	Kohl rabi	,, ...	Sept. ...	,, ,,
Chilo pyrocaustalis, Hmps.	*Sorghum vulgare* ...	Stem-borer	Aug. ...	Daura, ,, Kafin Soli, larvae
Ephestia cautella, Wlk.	*Gossypium hirsutum*	Stored seed	July ...	Kontagora larvae
Hellula undalis, F. ...	Cabbage	Roots ...	Feb. ...	Ibadan, larvae
	Turnip	Leaves ...	Oct. ...	,, ,,
Hymenia fascialis, Cram.	Beetroot	,, ...	,, ...	,, ,,
Loxostege fulvalis, Warr.	,, ...	,, ...	July ...	,, ,,
Plodia interpunctella, Hb.	*Vigna unguiculata* ...	Stored seed	Aug. ...	,, ,,
Pyrausta ferrugalis, F.	*Sterculia tragacantha*	Leaves ...	May ...	,, ,,
Zebronia phenice, Cram.	*Newbouldia laevis* ...	,, ...	Jan. ...	,, ,,

Name	Food-plants	Part attacked	Month of appearance	Locality and remarks
	RHYNCHOTA.			
Pentatomidae—				
Aspavia armigera, F.	*Vigna unguiculata*	Pods	Jan.	Ibadan, nymphs
	Oryza sativa	Seeds	Nov.	Igbogun, adults
A. hastator, F.	„ „	„	„	„ „
	Eleusine indica	„	May	Ibadan, „
Halyomorpha reflexa, Sign.	*Theobroma cacao*	Pods	Aug.	Owena, „
H. annulicornis, Sign.	*Vigna unguiculata*	„	Dec.	Ibadan, nymphs
Hotea subfasciata, Westw.	*Abutilon mauritianum*	Fruit	Feb.	„ adults and nymphs
Nezara viridula, L.	*Vigna unguiculata*	Pods	Jan.	Ibadan, nymphs
Piezosternum fallax, F.	*Solanum verbascifolium*	Fruit	June	Owena, adults
Plataspidae—				
Coptosoma marginella, Dall.	*Soja max*	Stems	Aug.	Ibadan, adults
Coreidae—				
Anoplocnemis curvipes, F.	*Vigna unguiculata*	Pods	Jan.	„ „
	Zinnia	Peduncles	Nov.	„ „
	Canavalia ensiformis	Shoots	Jan.	„ nymphs
Acanthocoris dentatus, Hagl.	*Theobroma cacao*	Pods	Aug.	Owena, adults
Acanthomyia horrida, Germ.	*Vigna unguiculata*	„	Jan.	Ibadan, nymphs
A. tomentosicollis, Stål	„ „	„	„	„ „
Cletomorpha lancigera, F.	*Theobroma cacao*	Leaves	Aug.	Owena, adults
C. fuscescens, Wlk.	*Solanum verbasifolium*	Fruit	July	„ „
Leptocorisa apicalis, Westw.	*Oryza sativa*	Seeds	Nov.	Igbogun, „
Mirperus torridus, Westw.	„ „	„	„	„ „
Riptortus dentipes, F.	„ „	„	?	Calabar „
	Vigna unguiculata	Pods	Dec.	Ibadan, „
Theraptus devastans, Dist.	*Mangifera indica*	Young leaves	Dec.	„ „
Lygaeidae—				
Aphanus littoralis, Dist.	*Arachis hypogaea*	Stored nuts	April	Ibi and Yola, adults and nymphs
Dieuches armipes, F.	*Solanum verbascifolium*	Fruit	July	Owena, adults
Graptostethus servus, F.	*Corchorus capsularis*	Seeds	Aug.	Ibadan, adults and nymphs
	C. olitorius	„	„	„ „
Oxycarenus dudgeoni, Dist.	*C. capsularis*	„	„	Ibadan, adults
O. gossypinus, Dist.	„ „	„	„	„ „ and nymphs
Pyrrhocoridae—				
Cenaeus gowdeyi, Bergr.	*Solanum verbascifolium*	Fruit	June	Owena, adults
Dysdercus superstitiosus, F.	*Mangifera indica*	„	April	Ibadan, „

Name	Food-plants	Part attacked	Month of appearance	Locality and remarks
	RHYNCHOTA—*continued.*			
CAPSIDAE—				
Halticus tibialis, Reut.	*Soja max*	Leaves ...	Aug. ...	Ibadan, adults
Helopeltis bergrothi, Reut.	*Theobroma cacao* ...	Pods and shoots	,, ...	Owena, adults and nymphs
	Solanum verbascifolium	Leaves ...	,, ...	,, ,,
H. westwoodi, White	,, ,,	,, ...	,, ...	Owena, adults
	Theobroma cacao ...	Pods ...	,, ...	,, ,,
Lygus neavei, Popp.	*Alchornea cordifolia*	Leaves ...	Dec. ...	Ibadan, ,,
Proboscidocoris fuliginosus, Reut. ...	*Vigna unguiculata* ...	Pods ...	,, ...	,, ,,
CERCOPIDAE—				
Locris maculata, F. ...	*Oryza sativa* ...	Leaves ...	Oct. ...	Benin, ,,
Ptyelus grossus, F. ...	*Acacia farnesiana* ...	Stem ...	July ...	Ibadan, adults and nymphs
RICANIIDAE—				
Pochazia fasciata, F.	*Theobroma cacao* ...	Shoots ...	Aug. ...	Owena, adults
	COLEOPTERA			
LUCANIDAE—				
Metopodontus savagei, Hope	*Solanum verbascifolium*	Fruit ...	July ...	,, ,,
DYNASTIDAE—				
Oryctes monoceros, Oliv.	*Phoenix dactylifera*...	Crown ...	Jan. ...	Fika, ,,
CETONIIDAE				
Heterorrhina africana, Drury	*Anacardium occidentale*	Fruits ...	April ...	Ibadan ,,
H. suturalis, F.	,, ,,	Fruit ...	March ...	,, ,,
Pachnoda cordata, Drury	,, ,,	,, ...	,, ...	,, ,,
Plaesiorrhina recurva, F.	,, ,,	,, ...	,, ...	,, ,,
TENEBRIONIDAE—				
Alphitobius laevigatus, F.	*Gossypium hirsutum*	Stored seed...	Nov. ...	Soba, larvae
ANOBIIDAE—				
Lasioderma serricorne, F.	Carrot	,, ,, ...	Jan. ...	Ibadan, adults
Stegobium paniceum, L.	*Solanum melongena*	,, ,, ...	Dec. ...	,, ,,
BOSTRYCHIDAE—				
Dinoderus minutus, F.	*Manihot* spp. ...	Stored Cassava starch	May ...	Lagos, larvae
COCCINELLIDAE—				
Epilachna similis, Thnb., var. *assimilis*, Muls.	*Sorghum vulgare* ..	Young leaves	June ...	Ibadan, adults

Name	Food-plants	Part attacked	Month of appearance	Locality and remarks
	COLEOPTERA—*continued.*			
Bruchidae—				
Pachymerus longus, Pic	*Arachis hypogaea*	Stored pods and nuts	Sept.	Adamawa Province, larvae
	Prosopis africana	Seed	,,	
	Tamarindus indica	Pods	,,	,, ,,
P. lacerdae, Chev.	*Elaeis guineënsis*	Fruit	June	Benin, larvae
Bruchus phaseoli, Gyll.	*Voandzeia subterranea*	Stored nuts	Jan.	Gamadio, ,,
B. obtectus, Say	Canadian Wonder beans	,, beans	July	Zaria, ,,
Lagriidae—				
Lagria viridipennis, F.	*Ricinus communis*	Leaves	,,	Ibadan, adults
Galerucidae—				
Barombia humeralis, Laboiss.	*Soja max*	Young leaves	Aug.	,, ,,
Ootheca mutabilis, Sahlb.	,, ,,	Leaves	,,	Yandev, ,,
Monolepta lineata, Karsch	French beans	,,	March	Ibadan, ,,
Halticidae—				
Podagrica spadicea, Dalm.	*Urena lobata*	,,	June	,, ,,
	Hibiscus cannabinus	,,	,,	,, ,,
P. uniforma, Jac.	,, ,,	,,	,,	,, ,,
	Urena lobata	,,	,,	,, ,,
Anthribidae—				
Phloeobius catenatus insignis, Jord.	*Theobroma cacao*	Stems of seedlings	Jan.	,, larvae
Scolytidae—				
Coccotrypes pygmaeus, Eich.	,, ,,	,, ,, ,,	Dec.	,, ,,
Curculionidae—				
Cylas puncticollis, Boh.	*Ipomoea batatas*	Stored tubers	Nov.	Bauchi, ,,
Isaniris thomsoni, Fst.	*Theobroma cacao*	Young shoots	August	Owena, adults
Piezotrachelus pullus, Boh.	*Vigna unguiculata*	Seeds	Dec.	Ibadan, larvae
Temnoschoita quadripustulata, F.	*Elaeis guineënsis*	Seedlings	Jan.	Warri, ,,
	DIPTERA			
Muscidae—				
Atherigona excisa, Thoms.	Cabbage	Stalk	Nov.	Ibadan, larvae
	Tomato	Fruit	June	,, ,,
	THYSANOPTERA			
Thripidae—				
Selenothrips rubricinctus, Giard	*Mangifera indica*	Leaves	Jan.	Ibadan, adults and nymphs
	ORTHOPTERA			
Gryllidae—				
Gryllotalpa africana, P. de B.	*Oryza sativa*	Seedlings	May	Ilesha, adults and nymphs

Name	Food-plants	Part attacked	Month of appearance	Locality and remarks
	ORTHOPTERA—*continued.*			
Acrididae—				
Catantops spissus, Wlk.	*Theobroma cacao* ...	Leaves ...	July ...	Ibadan, adults
	Aspilia latifolia ...	,, ...	Jan. ...	,, nymph
Cyrtacanthacris aeruginosa unicolor, Uv.	*Solanum melongena*	,, ...	Feb. ...	,, ,,
Heteropternis thoracica, Wlk.	*Pennisetum typhoides*	,, ...	Sept. ...	Nahuce, adults
Hieroglyphus daganensis, Kr.	,, ,,	,, ...	,, ...	,, ,, and nymphs
Kraussella amabile, Kr.	,, ,,	,, ...	,, ...	Nahuce, adults
Thisoicetrus leani, Uv.	,, ,,	,, ...	,, ...	,, ,, and nymphs
Kraussaria angulifera, Kr.	,, ,,	,, ...	,, ...	Gaya, adults and nymphs
Rhytidacris tectifera, Karsch	*Citrus paradisi* ...	,, ...	Feb. ...	Ibadan, adults
Zonocerus variegatus, L.	*Soja max*	,, ...	Nov. ...	,, nymphs
	Solanum verbascifolium	,, ...	Dec. ...	Owena, ,,

OBSERVATIONS ON MORTALITY AMONG INSECTS EXPOSED TO DRY INSECTICIDAL FILMS.

By J. R. Busvine, B.Sc., Ph.D., and Sarah Barnes, M.Sc., Ph.D.

Entomology Department, London School of Hygiene and Tropical Medicine.

Introduction.

Insecticide testing in the laboratory aims at precise discrimination between the insecticidal powers of different substances. To achieve accuracy it may be necessary to test under rather artificial conditions; for example at dosages much lower than would be employed in the field. Nevertheless it is clearly desirable to keep the conditions as close as possible to those obtaining in practice.

Hitherto, most widely used domestic insecticides have been used as solutions in refined kerosene sprayed directly at the insects by atomisers of the "flit gun" type. For testing this kind of insecticide, methods were developed in which the samples were dissolved in mineral oil and allowed to deposit on the insects in the form of a mist produced by special atomising guns. The Peet-Grady Chamber and its variants, the Campbell Turntable and the Potter Tower are examples of this method.

With the advent of permanent synthetic insecticides such as DDT, there has been a tendency to rely more on killing insects by residual films than by transient mists and sprays. Consequently the test methods need to be revised, since the insects may come into contact with dry films of crystals rather than drops of solutions.

As a preliminary sorting test the following method has been employed :—

Filter papers are impregnated with solutions of the insecticide calculated to give standard deposits per unit area. The volatile solvent is rapidly evaporated to leave a concentrated deposit which may be either an oil (pyrethrum concentrate), a crystalline solid or a supercooled liquid. The heavier deposits of DDT and Gammexane are certainly crystalline; the state of the lighter deposits is uncertain.

By this method it is possible to secure a reasonably uniform *lateral* distribution of toxicant (that can be shown by dyes or by insecticidal tests on different parts of the filter paper). But there is some doubt about the vertical distribution in the thickness of the filter paper that may depend on the conditions of evaporation of the solvent. An impermeable surface such as glass or metal would be more satisfactory from this point of view, but such surfaces provide no foothold for insects, some of which then tend to fall on their backs and thereby get an abnormal exposure. Consequently filter papers were taken as a conveniently available standard surface.

Details of the Method.

(a) *Preparation of test papers.*

Watkin's No. 1 filter papers 9 cm. in diameter were used. One cubic centimetre of acetone was found sufficient to soak a single paper. Solutions of various insecticides were prepared so that 1 cc. would give a deposit on the filter paper of 10, 5, 3, 1, ·1, ·01, ·001 or ·0001 mgm. per square centimetre. (The lower rates were prepared by 1 : 10 dilutions.) For impregnation, a paper was balanced on pin points and the liquid added spirally from a 1 cc. hypodermic syringe. The paper was fanned to accelerate the first stages of drying and this left the paper apparently dry in a minute or so. Further drying was allowed to continue for a few hours, before exposure of the insects.

(*b*) *Exposure of the insects.*

Walking insects such as lice, bugs and beetles, were confined on the papers in glass rings. Flying insects were confined between two petri dishes each lined with a filter paper. Where there was the possibility of a toxic vapour accumulating (Gammexane), the papers were placed face to face about 2 cm. apart and the intervening space enclosed by a ring of mosquito netting round the periphery.

When it was desired to keep the insects stationary, they were placed at the bottom of open glass tubes, one insect per tube, standing on the treated filter paper. Each tube was of sufficient bore to allow the insect to rest on the paper yet to prevent it moving in any direction. When it was required (in certain experiments) to keep the insects in more or less uniform movement, the insecticide-treated paper was impregnated further with a slowly volatilising irritant, namely 4-chlor 2-methyl phenol dissolved in benzene (described by Robinson, 1943, *Bull. ent. Res.*, **34,** pp. 269–277), to give a deposit of 0·0025 mg. of solute per sq. cm. (It had been previously ascertained that this dose of irritant was sufficient to keep the insects moving frequently without injuring them.)

(*c*) *Estimation of Results.*

For valid comparisons of different poisons it is necessary to determine equitoxic doses or exposure times, usually those for 50 per cent. kill. With contact insecticides sprayed directly on to the insects, the dose is a function of the concentration and the amount contaminating the insect. If the latter is kept constant, there is normally a linear relation between the mortality (expressed in probits) and the logarithm of the concentration. From this regression line, the median lethal concentration can be easily calculated.

With insects exposed to different deposits of *undiluted* toxicant it is not certain that a similar relation will hold. Two assumptions must be made: first, that the number of particles picked up by the insect is directly proportional to the number per unit area; second, that the toxic effect is related to the logarithm of the number of particles on the insect.

Owing to the inaccuracies in the method described (principally the unknown and possibly variable vertical distribution of insecticides in different papers) this toxic relationship cannot be accurately tested, but with the data available the method seems to give a satisfactory way of collating the results. In Section I of the results, therefore, the various substances will be compared by the rates of deposit estimated visually from log. deposit-probit regression lines to give 50 per cent. kill of different insects.

Section II of the results deals with mortalities caused by different periods of exposure on the treated filter papers. Insects walking on dry toxic films are exposed to intoxication in a peculiar way—presumably by the insecticide accumulating on the tarsi, legs and ventral surface (if this touches the substrate). Hence it is not surprising that some of the results are anomalous. The individual results are recorded since we have not discovered a comprehensive relationship between exposure time and mortality.

Insects and Materials Employed.

The following insects were used in the experiments :—

ORTHOPTERA	*Blatta orientalis*, L. (adults).
HEMIPTERA	*Cimex lectularius*, L. (adults).
	Rhodnius prolixus, Stål (4th and 5th nymphs and adults).
ANOPLURA	*Pediculus humanus*, L. (adults).

COLEOPTERA	*Tenebrio molitor*, L. (larvae). *Calandra granaria*, L. (adults). *Tribolium confusum*, Duval (adults).
DIPTERA	*Musca domestica*, L. (adults). *Aëdes aegypti*, L. (adults).

Also the following ticks :

IXODIDAE	*Ixodes ricinus*, L. (nymphs and adults).
ARGASIDAE	*Ornithodoros moubata*, Murray (nymphs).

The insecticides used were as follows :

DDT.—A sample of pure para-para DDT (m.p. 108°C.) was used. It was made at the Chemical Defence Experimental Station, Porton.

Gammexane.—Some pure *gamma* benzene hexachloride (m.p. 112°C.) was obtained from Imperial Chemical Industries Ltd.

Pyrethrum.—A concentrate containing 20 per cent. pyrethrins prepared by Messrs. S. B. Pennick and Co., of New Jersey, was used.

Rotenone.—Some crystalline rotenone obtained from Messrs. Stafford Allen was employed.

The experiments are grouped in two sections dealing with two different aspects of the problem, which we investigated more or less independently. (Section I records tests by J.R.B. and Section II tests by S.B.)

Results—Section I.

The following data give a comparison of the toxicity of the different insecticides to various insects with a constant period of exposure (two hours). The temperature during the exposure was maintained at 30°C. in order to stimulate the insects to a degree of activity.

(*a*) *DDT* :—

TABLE I.

Mortality of insects exposed to DDT films.

Insect	Average number of insects per test	Percent. kill of insects							
		Deposits : mg./sq. cm.							
		10	5	3	1	·1	·01	·001	·0001
Blatta	16	89	47	9	0	—	—	—	—
Cimex	25	96	50	21	11	—	—	—	—
Rhodnius	20	50	—	0	0	—	—	—	—
Pediculus	20	100	95	84	16	—	—	—	—
Tenebrio	16	55	—	0	0	—	—	—	—
Calandra	39	91	—	42	5	0	—	—	—
Tribolium	50	86	—	20	0	0	—	—	—
Musca	42	—	—	—	100	100	97	41	—
Aëdes	56	—	—	—	—	100	100	95	53
Ixodes	81	—	—	—	—	100	99	69	—
Ornithodoros	7	25	—	—	0	—	—	—	—

(*b*) *Gammexane* :—

TABLE II.

Mortalities of insects exposed to Gammexane films.

Insect	Average number of insects per test	Percent. kill of insects					
		Deposits : mg./sq. cm.					
		1	·1	·01	·001	·0001	·00001
Blatta	10	—	91	64	0	—	—
Cimex	37	—	—	98	59	5	—
Rhodnius	17	—	—	83	35	10	—
Pediculus	19	—	—	—	100	96	42
Tenebrio	14	8	6	—	—	—	—
Tribolium	59	—	100	58	4	—	—
Musca	35	—	—	—	100	100	100
Aëdes	20	—	—	—	100	100	100
Ornithodoros	9	—	100	90	11	—	—

(*c*) *Pyrethrins* :—

TABLE III.

Mortalities of insects exposed to Pyrethrin films.

Insect	Average number of insects per test	Percent. kill of insects					
		Deposits : mg./sq. cm.					
		1	·3	·1	·01	·001	·0001
Blatta	13	100	50	14	0	—	—
Cimex	18	—	—	100	87	32	—
Rhodnius	19	—	—	100	54	16	—
Pediculus	31	85	38	11	0	—	—
Tenebrio	16	50	8	—	—	—	—
Calandra	32	83	46	5	0	—	—
Tribolium	36	87	37	0	0	—	—
Musca	59	100	75	55	32	—	—
Aëdes	91	—	—	—	100	67	11
Ixodes	25	—	—	100	95	50	—
Ornithodoros	5	—	—	100	80	0	—

(*d*) *Rotenone* :—

TABLE IV.

Mortalities of insects exposed to films of Rotenone.

Insect	Average number of insects per test	Percent. kill of insects		
		Deposits : mg./sq. cm.		
		10	3	1
Blatta	10	0	—	—
Cimex	9	0	—	—
Rhodnius	8	0	—	—
Pediculus	8	0	—	—
Tenebrio	10	0	—	—
Calandra	20	0	—	—
Tribolium	20	0	—	—
Musca	38	100	67	25
Aëdes	25	6	25	5

From these data, the deposits expected to give 50 per cent. kills have been estimated and these figures are assembled in Table V.

TABLE V.

Median lethal deposits.

Insect	Deposit giving 50 per cent. kill after 2 hour exposure at 30°C.			
	DDT	Gammexane	Pyrethrins	Rotenone
Tenebrio	10	1	1	>10
Blatta	5	·005	·3	>10
Tribolium	5	·009	·4	>10
Calandra	3	—	·4	>10
Pediculus	2	·00001	·5	>10
Rhodnius	10	·002	·009	>10
Cimex	5	·0006	·002	>10
Ornithodoros	10	·003	·003	—
Musca	·001	·00001	·08	2
Aëdes	·0001	·00001	·0006	>10
Ixodes	·0005	—	·001	—

These results can be considered either in relation to the susceptibility of the insects, or the toxicity of the insecticides.

A. *Relative susceptibility of the test insects.*

It will be seen that, in Table V, the various insects and ticks have been arranged in groups according to their susceptibility to different insecticides. Thus :—

1. *Tenebrio* larvae are resistant to all the insecticides.

2a. *Blatta, Tribolium* and *Calandra* are resistant to DDT and Pyrethrins but susceptible to Gammexane.

2b. *Pediculus* is similar but much more susceptible to Gammexane.

3. *Rhodnius*, *Cimex* and *Ornithodoros* are resistant to DDT but susceptible to Gammexane and Pyrethrins.

4. *Musca* and *Aëdes* are very susceptible to Gammexane, pyrethrins and DDT. *Ixodes* is also sensitive to DDT and pyrethrins.

B. *Relative toxicity of insecticides.*

In spite of some irregularities from Table V, it would appear that the four insecticides show different degrees of toxicity ; Gammexane is most toxic, followed by pyrethrins, DDT and rotenone. However, it is important to realise that this is only valid for this type of experiment with dry films. In Table VI the equitoxic deposits are compared with median lethal doses obtained with the same insecticides applied in oil solutions ; it will be seen that the relationships are quite different as regards magnitude and even the order of toxicity. A possible partial explanation may be in the different physical states of the various substances. For example, the rotenone sample formed a hard glossy surface with the heavier doses (possibly owing to traces of resin) which may have impeded contamination of the insects. The actual physical states of the lower doses of DDT and Gammexane is difficult to determine with filter papers. On glass, an acetone solution of DDT dries to leave a supercooled liquid which crystallises out fairly rapidly, especially if stimulated mechanically.

TABLE VI.

Comparison of equitoxic doses of insecticides by different test methods. For the oil spray method the figures represent milligrams of toxicant per kilogram body weight. (Data for *Musca* and *Aëdes* from David, 1946*; data for *Cimex* and *Pediculus* from Busvine, 1946†). The figures for the dry films are taken from Table V.

Insect	Test method	Doses for 50 per cent. kill		
		DDT	Pyrethrins	Gammexane
Cimex	Oil spray ...	63	5	6
	Dry film ...	5	·002	·0006
Pediculus	Oil spray ...	27	42	1·5
	Dry film ...	2	·5	·00001
Musca	Oil spray ...	6–9	31–38	2–3
	Dry film ...	·001	·08	·00001
Aëdes	Oil spray ...	5–8	·5–1	3–3·5
	Dry film ...	·0001	·0006	·00001

*David (1946). Ann. appl. Biol., **33,** pp. 133–141. †Busvine (1946). Ann. appl. Biol., **33,** pp. 271–279.

Another point which must be remembered in interpreting these results is that they all refer to freshly prepared films. If the treated filter papers are stored, there are losses of toxicity which depend on the insecticide and the conditions of storage. The tests summarised in Table VII show that, whereas the lethal dose of DDT retains its power for at least three weeks, that of pyrethrum falls off gradually and the very low lethal doses of Gammexane quickly lose their effect. The failure of pyrethrins can be retarded by storage in darkness since daylight accelerates their chemical decomposition. The loss of power of Gammexane appears to be due simply to evaporation of the very sparse toxic films since their power can be preserved by storage in a desiccator over excess of Gammexane.

TABLE VII.

Ageing of insecticide films under different conditions. Except where stated to the contrary, the conditions of storage were at room temperature, exposed to light and air.

Test insect	Insecticide	Rate mgm. per sq. cm.	Conditions of storage	Percent. kill of insects by films of different ages			
				1 day	4 days	2 weeks	3 weeks
Pediculus ...	DDT	5		87	75	100	75
	Gammexane ...	·001		100	12	0	—
	Gammexane ...	·001	Desiccator with Gammexane	100	100	100	100
	Pyrethrum ...	1		87	26	43	0
	Pyrethrum ...	1	Darkness ...	85	87	57	0
Cimex	DDT	10		100	100	100	100
	Gammexane ...	·01		98	12	0	—
	Gammexane ...	·01	Desiccator with Gammexane	100	100	100	100
	Pyrethrins ...	0·1		100	50	33	25
	Pyrethrins ...	0·1	Darkness ...	100	100	100	37

Results—Section II.

The following data concern the influence on mortality of different periods of exposure having regard to the degree of activity of the insects; and also, continuous, as against intermittent exposure. These tests were conducted at room temperature.

(a) *Bed bugs, moving and stationary, on dry films of DDT and Gammexane.*

The percentage mortalities obtained among both stationary and moving bugs after different periods of exposure to different DDT deposits are summarised in Table VIII. Control series, in which bugs were exposed for the relevant periods, either stationary on clean filter papers or moving on filter papers impregnated with irritant only, showed that the percentage mortality among both groups of bugs was negligible, the highest being 2 per cent. after seven days' exposure.

TABLE VIII.

Percent. mortality among bugs both stationary and moving exposed for different periods to different deposits of DDT. (Average number of bugs from among which each percentage mortality was obtained was 58.)

DDT deposit (mg./cm.^{2})	5		1		0·5	
Time of exposure	Stationary	Moving	Stationary	Moving	Stationary	Moving
5 minutes	8	18	3	5	—	—
15 minutes	23	23	7	11	0	0
30 minutes	27	37	8	13	8	10
45 minutes	27	65	11	16	7	11
1 hour	57	82	20	33	23	31
2 hours	42	68	13	24	15	22
6 hours	50	78	19	25	17	20
18 hours	66	85	19	42	15	30
1 day	53	70	17	32	17	23
4 days	98	100	40	83	46	63
7 days	100	—	100	100	98	100

It is seen that :—

1. Percentage mortality among bugs kept moving over a given deposit of DDT for a given interval was generally higher than that among bugs kept stationary on the same deposit for the same period.

2. For each DDT deposit, the percentage mortality among bugs, whether stationary or moving, increased as the time of exposure increased from five to forty-five minutes. Among both classes of bugs exposed for periods ranging from one to twenty-four hours no such correlation was evident; indeed, the difference between percentage kill among bugs exposed for one hour and those exposed for twenty-four hours was negligible, and only after four days' exposure to the heaviest deposit, and seven days to the lighter ones, was a complete kill achieved.

This time lag in securing complete bug mortality on DDT films was surprising, and in order to ascertain whether or not a similar phenomenon occurred with any other insecticide, the trials were repeated on filter papers impregnated with deposits of gammexane ranging from 0·001 to 0·1 mg. per sq. cm. Since the lag occurred among both stationary and moving bugs alike on DDT films, these further trials were confined to stationary bugs only. The results summarised in Table XI, show that, for a given deposit, percentage mortality among bugs exposed to Gammexane generally increases as the time of exposure is increased.

TABLE IX.

Percent. mortality among stationary bugs exposed for different periods to different deposits of Gammexane. (Average number of bugs from which each percentage was calculated was 44.)

Deposit of gammexane (mg./cm.^{2})	·001	·01	·1
Time of exposure : ½ hour	12	28	36
1 hour	11	17	47
2 hours	79	100	100
6 hours	96	100	100
24 hours	100	100	100

(*b*) *Lice and flour beetles on dry DDT films.*

The question then arose whether or not this same phenomenon occurred among other insects exposed to DDT films. Lice and flour beetles were exposed for given periods of time on filter papers impregnated with deposits of either 0·5, 1 or 5 mg. of DDT per sq. cm. The percentage mortalities obtained among lice are summarised in Table X and those among beetles in Table XI.

TABLE X.

Percent. mortality among moving lice exposed for different periods to different deposits of DDT. (Average number of lice from which each percentage was calculated was 36.)

DDT deposit (mg./cm.^{2})	0·5	1	5
Time of exposure : ½ hour	0	0	5
1 hour	0	0	87
2 hours	37	59	100
6 hours	63	82	100
12 hours	100	100	100

TABLE XI.

Percent. mortality among moving beetles exposed for different periods to different deposits of DDT. (Average number of beetles from which each percentage was calculated was 49.)

DDT deposit (mg./cm.^{2})	0·5	1	5
Time of exposure : ½ hour	—	5	23
1 hour	—	8	34
1½ hours	—	11	41
2 hours	—	16	48
6 hours	11	24	76
18 hours	24	50	100
24 hours	28	56	100

From these tables it is seen that for a given deposit of DDT, percentage mortality among both test insects increased as the time of exposure was prolonged.

(c) *Mortality among bed bugs after intermittent exposure to dry DDT films.*

The tests described below were undertaken to compare mortality among bugs after intermittent exposure to DDT, such as would probably occur under field conditions, and that among bugs exposed continuously over a given period. Bugs were exposed for varying intervals on filter papers impregnated with a DDT deposit of 5 mg. per sq. cm. the total period of exposure ranging from one to six hours. The results obtained are summarised in Table XII.

TABLE XII.

Percent. mortality among bugs exposed intermittently to DDT films. (Average number of bugs from which each percentage was calculated was 39.)

Interval of exposure	Total period of exposure	Per cent. mortality
¼ hour	1 hour	75
½ hour	1 hour	88
½ hour	2 hours	76
1 hour	2 hours	70
½ hour	6 hours	74
1 hour	6 hour	79

Comparing these with the relevant figures in Table VIII, it is seen that intermittent exposure totalling a given period of time yields approximately the same percentage mortality as that obtained after continuous exposure for the same period. Moreover, the length of the intervals comprising the total exposure appears to have no influence on ultimate mortality.

Summary.

A variety of insects were exposed to films of insecticides by confining them for various periods on filter papers impregnated by a volatile solvent method. The following results were observed :—

1. Nine species of insect and two species of tick were exposed to films of different insecticides for a standard exposure of two hours at 30°C. It was found that freshly prepared films of Gammexane were most toxic, followed by pyrethrins, DDT and rotenone.

2. The relative toxicity of the films of undiluted insecticides is considerably different from that of the same insecticides applied in oily solution.

3. The very sparse toxic films of Gammexane soon lost their toxicity, apparently by evaporation. The pyrethrin films were also less persistent than those of DDT.

4. The test organisms can be arranged in groups showing different types of susceptibility.

5. Percentage mortality among bugs kept in motion for a specified period over a given deposit of DDT is greater than that among stationary bugs exposed to the same deposit for the same period. The addition of an irritant to a DDT spray would therefore enhance the efficiency of the insecticide.

6. A marked time lag in achieving complete mortality among bugs exposed to dry DDT films is recorded. No such lag was observed among bugs exposed to Gammexane nor among lice and flour beetles exposed to DDT. In these latter cases, percentage mortality gradually increased as the time of the exposure increased.

7. Percentage mortality among bugs exposed intermittently to DDT for a given total period of exposure did not differ appreciably from that obtained among bugs exposed continuously for the same period. After intermittent exposure for a particular total time, the length of the intervals during which the insects were in contact with the insecticide did not influence the percentage mortality.

ON SOME SANDFLIES (*PHLEBOTOMUS*) OF THE *SERGENTI* GROUP IN PALESTINE.

By O. Theodor.

Department of Parasitology, Hebrew University, Jerusalem.

The distribution of *Phlebotomus sergenti* in Palestine, Syria and Mesopotamia has received special attention, because of the rôle of this species in the transmission of oriental sore.

Sinton (1927), in discussing the distribution of *P. sergenti* and oriental sore, criticised the diagnosis of *P. papatasii* made by Adler and Theodor in 1925, in connection with transmission experiments carried out with sandflies caught in Jericho, and pointed out that a diagnosis based on negative evidence is not conclusive. He assumed the occurrence of *P. sergenti* in the Jordan valley on the authority of Larrousse (1924). This author, however, did not actually record *P. sergenti* from the Jordan valley. He showed in his note that *Phlebotomus el Ghor*, Schroetter, was not a *Phlebotomus* at all, but a species of *Psychoda* or *Pericoma*. He then states: " Les véritables Phlébotomes existent dans les régions mentionnées par H. von Schroetter en Syrie et en Mésopotamie. Divers auteurs y signalent les espèces suivantes : *P. papatasii* Scop., *P. minutus* Rond., *P. minutus* var. *africanus* Newstead et *P. sergenti* Parrot, au total trois espèces et une variétée."

This statement does not imply that *P. sergenti* occurs in Palestine, but that it occurs in some of the countries mentioned.

P. sergenti is the predominant species in certain parts of Baghdad, where it is responsible for the transmission of oriental sore. It is as common as *P. papatasii* in Aleppo where both species probably transmit the disease. In endemic foci of oriental sore in the Lebanon and in Palestine (Bar Elias, Jericho), no *P. sergenti* has been found, although thousands of sandflies were dissected and identified. The actual proof that *P. papatasii* transmits oriental sore has been given by Adler and Theodor (1929a) who re-examined a slide with a preparation of an infected sandfly that had been used for a successful inoculation experiment, and found it to be *P. papatasii* according to the pharyngeal armature. The final proof has been given by Adler and Ber (1944) in successful transmission experiments by the bite of infected *P. papatasii* in five out of nine cases.

Since 1926 *P. sergenti* has been found only very rarely in Syria (except Aleppo) and Egypt. The records of *P. sergenti* in these countries are as follows :—

Damascus, 1 ♂, 1 ♀. 6.vi.23 (old slide from collection).
Bcherreh (Lebanon), 1 ♀. 1928.
Maadi near Cairo, 1 ♀. 7.vii.43.

On 2nd December, 1946, a single female of *P. sergenti*, typical in every respect, was found in a cave in Wadi Fukra about 15 km. south of the south end of the Dead Sea. This is the first record of this species in Palestine. It is all the more interesting as several thousand sandflies have been examined from settlements near the North end of the Dead Sea, and from Jericho, without finding a single specimen of *P. sergenti*.

The relative rarity of this species in Syria (so far as our knowledge goes) makes it unlikely that it plays any rôle in the transmission of oriental sore in the endemic foci examined, except in Aleppo, where it is common.

Terminology.—As pointed out by Tonnoir (1935), the terminology used by the various authors for the description of the male genitalia of *Phlebotomus* is unsatisfactory as it does not comply with the terms accepted in entomological literature. Christophers and Barraud (1926) established the homology of the hypopygium of *Phlebotomus*. Their terminology, which is given below, will be used in the present paper :—

1st segment of the superior clasper = Coxite.
2nd segment of the superior clasper = Style.
Median appendages = Parameres.
Intromittent organ = Penis {penis filaments + penis sheath.
Inferior clasper = Lateral lobes.
Submedian lamellae = Cerci.

A few specimens of *Phlebotomus* were caught in Palestine in 1934 and 1945 which are related to *P. sergenti* and *P. alexandri*, but show sufficiently marked differences to justify the creation of new species.

Phlebotomus maris-mortui, sp. n.

During an excursion from Hebron to Engedi on the shore of the Dead Sea in May 1934, four female sandflies were caught biting members of the excursion which were sleeping under an overhanging rock in the Wadi el Ghor, 15 km. west of Engedi. A single male which apparently belongs to this species was caught in October 1946 in a cave in Ein Feshha near the north end of the Dead Sea, about 30 km. north of the place where the females had been caught previously.

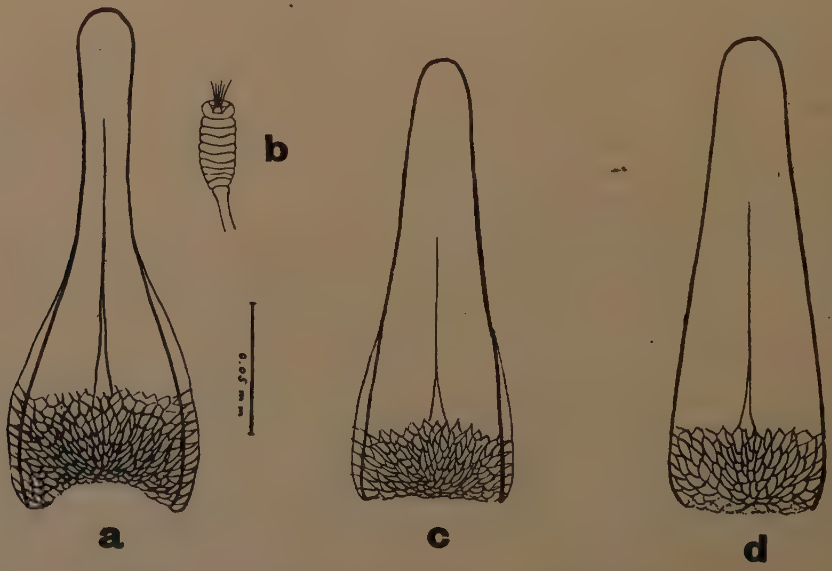

Fig. 1.—(*a*) Pharynx of female of *P. maris-mortui*, sp. n.; (*b*) Spermatheca of *P. maris-mortui*, sp. n.; (*c*) Pharynx of female of *P. alexandri* from Cyprus; (*d*) Pharynx of female of *P. alexandri* from India.

Female.

Size: 2·3 mm.

Wings: Length: 2·35 mm. Breadth: 0·6 mm. Wing index $\frac{\alpha}{\beta}$=1·4 to 1·6.

Antennae: Segment 3=4+5. Length of segment 3=0·16 mm. In one specimen segment 3 is slightly longer than 4+5, in two others slightly less. $\frac{A3}{E}$=0·5. Ascoids (geniculated spines) rather long, reaching end of segment.

Palps: Formula 1, 4, 2, 3, 5. Relative length of segments: 1, 3, 3·8, 2·4, 6.

Pharynx: A strong armature of medium sized teeth, of which the outer are inclined sidewards. The teeth are smaller than in *P. sergenti* and about as big as in *P. alexandri*. Viewed dorsally the teeth of the dorsal plate seem to extend laterally beyond those of the ventral plates (fig. 1 *a*). The posterior margin of the pharyngeal armature is markedly concave.

Spermathecae: 8 or 9 segments. The apical segment is rounded and wider than the other segments (fig. 1 *b*).

Male.

Size: 1·85 mm.

Wings: Length: 1·6 mm. Breadth: 0·4 mm. Wing index $\frac{\alpha}{\beta}$=1·6.

Antennae: Segment 3<4+5. Length of segment 3=0·12 mm. $\frac{A3}{E}$=0·75. Two ascoids on segments 3 to 15 which do not reach the end of the segment.

Palps: Formula 1, 4, 2, 3, 5. Relative length of segments: 1, 2·5, 3·5, 2·1, 6.

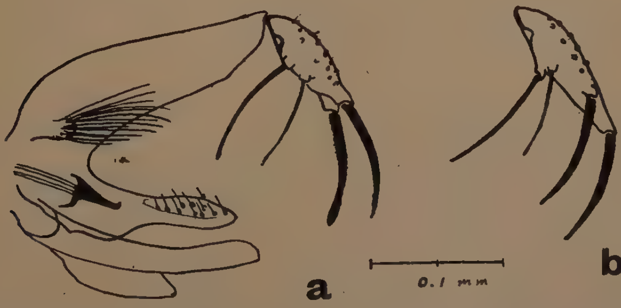

Fig. 2.—(*a*) Male genitalia of *P. maris-mortui*, sp. n.; (*b*) Style of *P. alexandri* from Cyprus.

Pharynx: Weakly developed armature of scale-like teeth.

Genitalia (fig. 2 *a*). Style shorter than half the coxite. Coxite=0·21 mm. Style=0·09 mm. There are four spines on the style of which two stout ones are terminal on moderately long pedicles, a thinner spine at the basal third of the segment and a still thinner one about halfway between the basal and the terminal spines. The process with hairs near the base of the coxite is rounded and short, similar to that of *P. alexandri*. Penis short and hooked (0·05 mm.). Lateral lobes of 9th tergite shorter than coxite (0·17 mm.). Parameres with flat upper surface which is covered with short hairs.

Material examined: 4 ♀♀. Wadi el Ghor near Dead Sea. May 1934. 1 ♂. Ein Feshha near Dead Sea. October 1946. In cave.

P. maris-mortui is closely related to *P. alexandri* which was described by Newstead (1920) as *P. sergenti* var. from Mesopotamia. Sinton (1928) called it *P. sergenti* var. *alexandri* and considered the Indian and Mesopotamian material identical. Parrot (1936) raised it to specific rank. An examination of specimens of *P. alexandri* from India has shown, however, that they differ from specimens caught by Adler in Cyprus (1946) and from material described by Parrot from North Africa (1936). In the Indian female specimens the teeth in the pharynx are bigger (fig. 1 *d*) and the ascoids of the antennae do not reach the ends of the segments as in specimens from Cyprus and Algeria. The palp formula of the Indian specimens is as follows:—

Male: 1, 2, 4, 3, 5. Relative length of segments: 1, 2·5, 3·6, 2·9, 6.

Female: 1, (2, 4) 3, 5. Relative length of segments: 1, 2·5, 3·6, 2·6, 5·2.

Antennae:

Male: Segment 3>4+5. $\frac{A3}{E}=0{\cdot}8$. Ascoids do not reach end of segments.

Female: Segment 3=4+5. $\frac{A3}{E}=0{\cdot}5$. Ascoids as in male.

Cyprus material. Palp formula.

Male: 1, (2,4) 3, 5. Relative length of segments: 1, 2·4, 3·2, 2·4, 5·5.

Female: 1, 4, 2, 3, 5. Relative length of segments: 1, 2·9, 3·5, 2·4, 5·2.

Antennae:

Male: Segment 3>4+5. $\frac{A3}{E}=0{\cdot}7$. Ascoids not reaching end of segments.

Female: Segment 3=4+5. $\frac{A3}{E}=0{\cdot}5$. Ascoids reaching or nearly reaching end of segments.

The palp formula given by Parrot for specimens from Algeria and the length of the ascoids is like that of the Cyprus specimens.

A male from Turkestan has the palp formula: 1, 4, 2, 3, 5. Relative length of segments: 1, 2·6, 3·3, 2·3, 5·2.

Antennae: 3=4+5. $\frac{A3}{E}=0{\cdot}7$. Ascoids not reaching end of segment.

The palp and antennal measurements of the Cyprus and Algerian material are constant in a sufficiently large series of specimens, but an examination of more material from India and Turkestan is necessary to determine whether the differences described are constant.

In all the forms of *P. alexandri* mentioned above, the wing is relatively short (1·6 to 1·8 mm. as against a body length of 2·2 to 2·4 mm.) and this constitutes one of the main differences of *P. alexandri* from other species of the *sergenti* group.

It seems clear that the Mediterranean form of *P. alexandri* differs from the Indian and possibly from the Turkestan form. I have not, however, been able to examine material from Mesopotamia and the relation of the Mediterranean and the Indian form to the type form cannot, therefore, be determined.

P. maris-mortui is closely related to *P. alexandri* but differs from all its various forms in the pharyngeal armature, which is wider and concave posteriorly, and in the relatively greater length of the wing which is as long as the body in *P. maris-mortui* and much shorter than the body in all the forms of *P. alexandri*. The single male of *P. maris-mortui* examined is a very small specimen and its wing is relatively shorter than that of the females and approaches the relation between body and wing length found in *P. alexandri*. The palp and antennal formulae of *P. maris-mortui* resemble those of *P. alexandri* from Cyprus. In the male genitalia the style is much shorter than in the various forms of *P. alexandri* and the distribution of the spines is different. In *P. alexandri* only one spine is terminal, the second one originates at the apical third and both basal spines are situated at about the same level at the basal third of the segment. The penis and the lateral lobes are also relatively shorter than in *P. alexandri*. The male genitalia of *P. maris-mortui* are rather similar to those of *P. sergenti* from which they differ in the form of the process at the base of the coxite and the position of the thinnest spine on the style. The third segment of the antenna is also much shorter than in *P. sergenti*.

Note.—Since the above was written, two more females of *P. maris-mortui* were caught on 19th November, 1946, in the same cave in which the male of this species was found a month earlier. Together with the two females, two males were caught which were found to be typical *P. alexandri*, a species which has not been recorded previously from Palestine.

The palp formula of these males is : 1, 2, 4, 3, 5. Relative length of segments : 1, 2·3, 3·5, 2·6, 6·2, *i.e.* the fourth segment is longer than the second, while both segments are equal in specimens from Cyprus and the fourth is shorter than the second in *P. maris-mortui*. The palp formula thus resembles that of the male specimen from India mentioned above. The wing length is 2 mm. and the size of the insects is 2·2 mm. The wing is thus slightly longer relatively than in the other forms of *P. alexandri* examined.

Phlebotomus jacusieli, n. sp.

A single male of this species was caught in Rosh Pinah in Northern Palestine in July 1945 by Major F. Jacusiel, R.A.M.C., to whom this species is dedicated.

Male.

Size : 2·1 mm.

Wings : Length : 2·2 mm. Breadth : 0·65 mm. Wing index $\frac{\alpha}{\beta}=1{\cdot}3$.

Antennae : Segment 3 slightly shorter than 4+5. $\frac{A3}{E}=1{\cdot}16$.

Palps : Formula 1, (2, 4) 3, 5. Relative length of segments : 1, 2·9, 3·9, 2·9, 7.

Pharynx : Armature rather weakly developed, appearing as a network of lines, similar to that of males of *P. sergenti*.

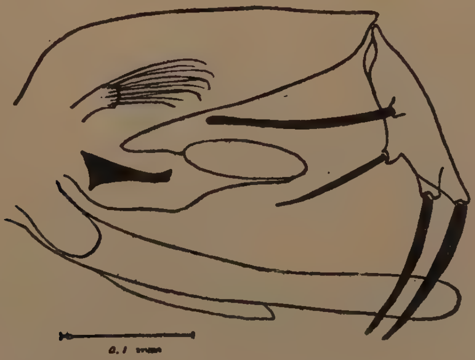

Fig. 3.—Male genitalia of *P. jacusieli*, sp. n.

Genitalia : Style slightly longer than half the coxite, with one spine terminal, a second distinctly subterminal on a long pedicle, a third weaker one at about half the length of the style and a stronger fourth spine at the basal quarter. Club shaped process at base of coxite long and slender as in *P. sergenti* with brush of long curved hairs. Penis with distinct terminal hook (fig. 3).

P. jacusieli resembles *P. sergenti* in all body measurements, but the style is much longer and resembles that of *P. alexandri*. The measurements of antennae, wing length and the form of the style are remarkably constant throughout the *sergenti* group and the conditions found in the specimen described above cannot be attributed to variation in any of the known species of the *sergenti* group. It is therefore considered a new species.

Phlebotomus palestinensis, Adler & Theodor, 1927.

This species was described from a single female caught in Jericho in December 1925. Subsequently, three females were caught in Baghdad in June 1928, one female in Abu Germ in Northern Persia in June 1929 and one female in Chugim at the Northern shore of the Dead Sea in June 1941. Among material collected in Jericho in July 1934, which was re-examined recently, a male was found, which clearly belongs to this species.

Male.

Size : 1·6 mm.

Wing : Length : 1·4 mm. Breadth : 0·26 mm. Wing index $\frac{\alpha}{\beta}=0{\cdot}4$.

Antennae : missing.

Palps : Formula : 1, 2, (3, 4) 5. Relative length of segments : 1, 3, 4·6, 4·6, 9.

Epipharynx : Length 0·13 mm.

Buccal cavity : Armature consisting of a straight row of 12 or 13 teeth with moderately long points. Pigmented area very faint, roughly triangular (fig. 4 *b*).

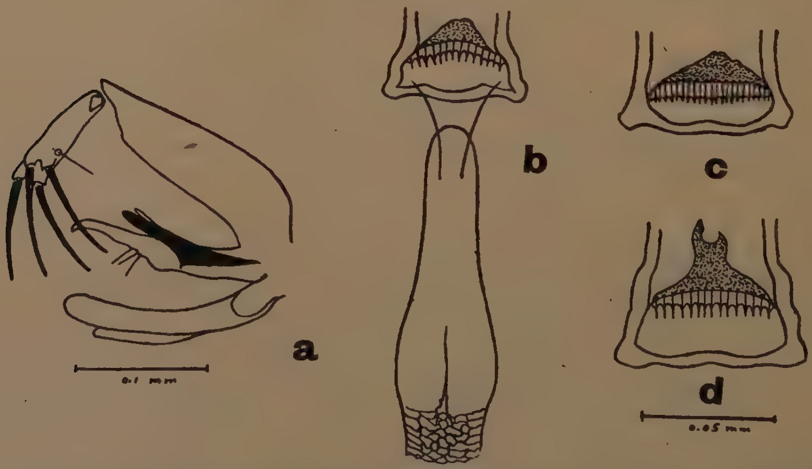

Fig. 4.—*P. palestinensis*. (*a*) Male genitalia. (*b*) Pharynx and buccal cavity of male. (*c*) Buccal cavity of female (type form). (*d*) Buccal cavity of form with 15 teeth and bifid pigmented area.

Pharynx : Twice as wide posteriorly as anteriorly. Armature consisting of several rows of irregular, blunt, well marked teeth (fig. 4 *b*).

Genitalia (fig. 4 *a*) : Style half the length of the coxite. Four spines, of which two are terminal and two subterminal. The small internal seta is situated at the basal quarter of the style. Parameres slender with hooked tip. Penis slender, tapering to a blunt rounded point. Lateral lobes as long as coxite.

Female.

The original description was rather short and some details from the five females caught subsequently are given herewith to complete the description.

Size: 1·8 to 2·2 mm.

Wing. Length: 1·6 mm. to 1·8 mm. Breadth: 0·4 to 0·45 mm. Wing index =0·5 to 0·6.

Palps: Formula: 1, 2, (3, 4,) 5. Relative length of segments: 1, 3, 5, 5, 9.

Antennae: Segment $3 < 4 + 5$. Ascoids rather short and stumpy.

Epipharynx very short and wide. $\frac{A3}{E} = 1{\cdot}1$ to $1{\cdot}3$.

Buccal cavity: Armature of 20 to 22 parallel teeth with moderately long points, standing in a straight row. A second row of short teeth at the base of the long ones (fig. 4 *c*).

In the specimen from Chugim near the Dead Sea and in one specimen from Baghdad there are only 15 to 16 teeth with much longer points and the anterior process of the pigmented area is distinctly bifid (fig. 4 *d*). In the specimen from Persia there are 20 teeth and the pigmented area is bifid with short processes. There seems, therefore, to be intergradation between the type form and the form with only 15 teeth and bifid pigmented area. More material will have to be examined in order to decide whether this form is a separate variety.

Pharynx: Twice as wide posteriorly as anteriorly with an armature of numerous, strongly marked, small teeth. Behind the main armature a number of fine teeth with very long points.

Spermathecae: Round or broadly elliptical capsules with rather wide ducts.

P. palestinensis is related to *P. sogdianus*, Parrot, which has, however, 33 to 35 teeth in the buccal cavity in the female and 22 to 24 in the male. The male genitalia of *P. palestinensis* are very similar to those of *P. sogdianus.*

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THE COTTON JASSID (*EMPOASCA LIBYCA*, BERG.) IN THE ANGLO-EGYPTIAN SUDAN, AND EXPERIMENTS ON ITS CONTROL.

By J. W. Cowland.

Senior Entomologist, Research Division, Department of Agriculture and Forests.

The cotton Jassid, *Empoasca libyca*, de Bergevin, was first suspected of being an important pest of cotton in the season 1934–35 in the recently canalised area of the northern Gezira. These insects had previously been numerous in some years in the Messellemia Group, and had probably caused some damage, but the colour changes due to normal maturation of the leaves had masked the symptoms. The damage caused has been increasing and the species is now a major pest in the northern and western blocks of the Gezira Irrigated Scheme.

Michelmore (1930) studied, at the Gezira Research Farm, Wad Medani, this Jassid which was then incorrectly determined as *Empoasca facialis*, Jacobi. At that time leaf curl, a virus disease of cotton, was prevalent and this insect was one of the possible transmitting agents, but Kirkpatrick (1930) proved that the disease was carried and transmitted solely by whitefly, *Bemisia gossypiperda*, M. & L.

Distribution of Jassids and their Food Plants in the Sudan.

Empoasca libyca, Berg.

This species was described from specimens taken on vines in Tripolitania where de Bergevin and Zanon (1922) reported it as causing serious damage.

Paoli (1932) described a species from Italian Somaliland, Tanganyika, and Anglo-Egyptian Sudan on cotton and figured the male genitalia, naming it *E. benedettoi*. Later (1936 b) he compared his specimens with co-types of de Bergevin's species and failed to trace any differences. He therefore sunk *E. benedettoi* as a synonym of *E. libyca*.

The species has a wide geographical range in Africa, occurring from the shores of the Mediterranean to as far south as Tanganyika. It is the only abundant *Empoasca* in the northern Sudan, north of latitude 10°N. Specimens have not been received from Equatoria Province, but it is probably present as it occurs in Uganda, where Hargreaves (1936) observed reddening of cotton leaves attacked by three species of Jassids, of which *E. libyca* was the most abundant.

E. libyca feeds on a large number of plants ; the adults and the nymphs have been bred to maturity on the following in the Sudan :—

Malvaceae :—Cotton, hollyhocks, *Hibiscus esculentus*, *Sida* sp., *Abutilon* spp.

Tiliaceae :—*Corchorus* spp.

Solanaceae :—Potato, tomato, egg-plant, *Solanum dubium*, *Datura* sp., *Withania somnifera*.

Leguminosae :—Egyptian bean, haricot bean.

Adults have also been taken on the following plants, but it is not definite whether they oviposit and complete their life-cycle on them :—

Leguminosae :—*Dolichos lablab*, *Cajanus cajan*, *Indigofera paucifolia*, *Rhynchosia memnonia*, *Medicago sativa*.

Pedaliaceae :—*Sesamum indicum*.

Graminaceae :—*Sorghum*.

Labiatae :—*Ocimum basilicum*.

Erythroneura lubiae, China.

E. lubiae was described by China (1931) from specimens collected from the Anglo-Egyptian Sudan. Its chief food plants belong to the Leguminosae, but it is also abundantly found on plants of other genera in association with *E. libyca,* from which it can be recognised by its larger size and greener colour, and by the wing veins, which in *Erythroneura* run to the distal margin of the wing, while in *Empoasca* they form a closed cell and do not run to the margin.

E. lubiae has been collected from the following plants :—

Leguminosae : *Dolichos lablab, Medicago sativa, Indigofera* sp., *Rhynchosia memnonia, Vigna* sp., *Canavalia* sp.

Malvaceae : *Hibiscus esculentus, Abutilon* sp., cotton.

Tiliaceae : *Corchorus* spp.

Solanaceae : *Withania somnifera, Solanum dubium.*

Vitaceae : Vine.

Labiatae : *Ocimum basilicum.*

Convolvulaceae : *Ipomoea cordofana.*

Also on Zinnia, Fennel and Canna.

It is widespread throughout the irrigated areas of the Sudan and in the Northern Province is abundant on American cotton, but in the Gezira area it only occurs occasionally on the Egyptian type cotton.

Empoasca facialis, Jacobi

This species was described by Jacobi (1912). From the examination of the male genitalia of the Jassids labelled *E. facialis* in the Sudan collection and of recent material received from the south, this species is now known to occur on rain grown cotton at Talodi, Kordofan Province, and at Meridi, Equatoria Province, on cotton and sweet potatoes.

Empoasca distinguenda, Paoli

This species was described by Paoli (1932). It is known from Africa south of the Equator and from the Transvaal, where it was taken on cotton and *Phaseolus acutifolius.* It occurs also in the Belgian Congo. Paoli (1936 a) records it on castor oil in Upper Egypt and the writer bred it on the same plant on the Gezira Research Farm, Wad Medani, in January 1946.

Empoasca dolichi, Paoli

This species was described by Paoli (1930) from specimens collected from *Dolichos lablab* in Italian Somaliland, and later it was reported as abundant on cotton and leguminous crops from Uganda, Transvaal, Nigeria and the Belgian Congo.

Russo (1935) reported it as injurious to cotton in Somaliland. It is a tropical species and has not been found as far north as the Gezira.

Damage caused by Empoasca libyca.

Smith and Poos (1931) have shown that Jassids can be divided into two groups. One group is characterised by the habit of feeding on the mesophyll tissue of the leaves, producing a definite stippling or spotting on the upper surface. The more mature foliage seems to be preferred to that of the more succulent leaves. *Erythroneura lubiae* belongs to this group.

The second group feeds by penetrating and plugging the phloem and xylem vessels, and the well-being of the group depends on the availability of succulent tissue. This type of feeding causes more serious injury to the plant, as it results in interference with the translocation of plant materials, causing symptoms known as "hopperburn". *Empoasca libyca* belongs to this group.

The first symptoms of hopperburn appear as a paling at the leaf edges. This rapidly extends to the tissue between the main veins, producing reddening and yellowing and eventual death of the leaf tissue from the periphery inwards, as if scorched. When the attack is heavy the young leaves fail to expand fully and remain small and curled with brown dry margins. Growth is brought to a premature standstill and the apical buds and young bolls are shed.

The following cage experiment, carried out in 1935, showed conclusively that Jassids were able to cause such symptoms.

In a field of Sakel Cotton, in Um Degarsi Block, twenty plants of similar growth were selected at the end of October and each was enclosed in a muslin cage 50 × 50 × 100 cms. with a sliding glass front. After caging, all plants were sprayed with nicotine sulphate and soap. After a fortnight (November 4) the plants were resprayed and in ten of the cages approximately 100 Jassid nymphs were introduced, while the remaining ten cages acted as controls.

After a fortnight some of the leaves in the infested cages began to show yellowing along the leaf margins extending between the main veins and a number of brown puncture marks. Finally the leaves dried up as if scorched, with only a narrow green margin along the main veins. Growth ceased and all the top bolls and buds were shed.

Plant diagrams were made fortnightly and in Table I below is given the average number of potential fruits per plant each fortnight on Jassid-free and Jassid-infested plants. The yields of each plant are given in Table II.

TABLE I.

Average number of potential fruits.

Date	Jassid-free Cages			Jassid-infested Cages		
	Buds	Bolls	Open bolls	Buds	Bolls	Open bolls
5.xi.35	15	·7		16·2	·2	
19.xi.35	20·4	7·1		14·3	5·5	
3.xii.35	21·8	10·0		15·1	9·2	
19.xii.35	18·1	18·0		7·1	15·9	
1.i.36	11·7	24·7	·7	1·3	15·7	
14.i.36	7·9	28·0	1·7	·3	14·7	1·0
29.i.36	3·6	26·9	4·8	·1	9·4	4·7
11.ii.36		25·7	3·7		4·4	4·9
25.ii.36		19·4	5·7		1·2	2·9
11.iii.36		10·0	10·3		·4	·9
7.iv.36		1·6	8·1			·4
Total bolls matured			35·0			14·8

TABLE II.

Yields

Jassid-free Cages				Jassid-infested Cages			
Plant No.	Open bolls per plant	Average weight seed cotton per boll	Yield per plant in grams	Plant No.	Open bolls per plant	Average weight in grams seed cotton	Yield per plant in grams
14	34	3·22	109·71	11	15	2·60	39·02
12	49	1·61	128·17	16	13	2·71	35·22
15	38	2·57	97·71	19	13	2·82	36·70
5	37	2·87	106·21	5	17	2·51	52·74
4	27	3·27	88·39	8	17	2·62	44·65
6	44	2·73	120·44	2	19	2·50	47·01
9	28	3·13	87·71	10	13	2·31	30·02
23	24	2·83	67·93	21	9	2·68	24·11
17	35	2·76	94·12	22	11	2·53	27·89
24	—	—	—	20	21	2·38	49·91
Total ...	316	24·99	900·39	—	148	25·66	387·27
Mean ...	35	2·89	100·04	—	14·8	2·57	38·73

Life-cycle of Empoasca libyca.

Michelmore (1930) found that the egg and nymphal stages were of short duration and followed by a long adult life in which the female lays her eggs over a long period. The eggs, of which four are usually ripe at one time in the female, are relatively large, cylindrical and slightly curved with rounded ends. They are laid embedded in the soft cortex of the leaf stems and main veins but not in the lamina, and are invisible from the outside. The young nymph is at first colourless but soon becomes a unicoloured green or yellowish-green. The egg and nymph stages last about eight days each and the adult as long as two months. In the writer's experiments the shortest period of the egg stage was 6 days with an average of 8 to 10 days. The nymph passes through 5 instars in the course of 8 to 12 days. The length of all immature stages increases during the colder dry weather of winter.

A number of experiments have been carried out to find out the reproductive potential of the adults during the cotton season. Cotton was sown on a small plot on the twelfth of August, September and October and individual plants were subsequently caged for periods long enough to allow any eggs on them to hatch (ten days in September and October). Each was then infested with five pairs of Jassids, which were left on as long as any of the females remained alive.

On 28th September five plants of the August sowing were thus infested and from these five cages an average of 205 nymphs were obtained with the largest number of 359 nymphs from one cage.

From a further five cages, similarly infested on 8th October, an average of 295 nymphs was obtained and a maximum from one cage of 395.

On 13th November from five plants of the August sowing and five plants of the September sowing, each infested with 5 pairs of Jassids, an average of 77 and 138 nymphs respectively was obtained with a maximum of 158 from one cage.

On the 8th January from five plants of the August, five plants of the September, and five plants of the October sowings each infested with 5 pairs of Jassids an average of 42, 57, and 13 nymphs respectively was obtained with a maximum of 80 from one of the September sown plants.

It is apparent from these experiments that Jassids are more prolific when climatic conditions favour plant growth.

Methods and Apparatus.

(a) *Sweeping.*—Verma and Afzal (1940) in India devised a reliable and quick method for estimating the comparative infestation of Jassids on different varieties. They showed by statistical methods that the level of significance was practically the same by sweeping, counting nymphs or a combination of both. The writer therefore decided to rely on adult counts by the sweeping method wherever possible and to supplement, if necessary, by nymphal counts.

Standard funnel-shaped calico nets were made, 2 feet in length and 1 foot in diameter at the base. These were attached to a wire ring which could be screwed on to a wooden handle 2 ft. 6 in. long. To take a sample the operator walked between the cotton-rows taking twelve forward and backward sweeps over two rows and repeated it over another two rows. (In some cases where the rows were short only 6 double sweeps were made instead of twelve.) The net was then tied up and removed to a tin containing cyanide to kill the insects ; a fresh net was attached and another sample taken until all subplots had been swept. Later the insects were counted in the laboratory.

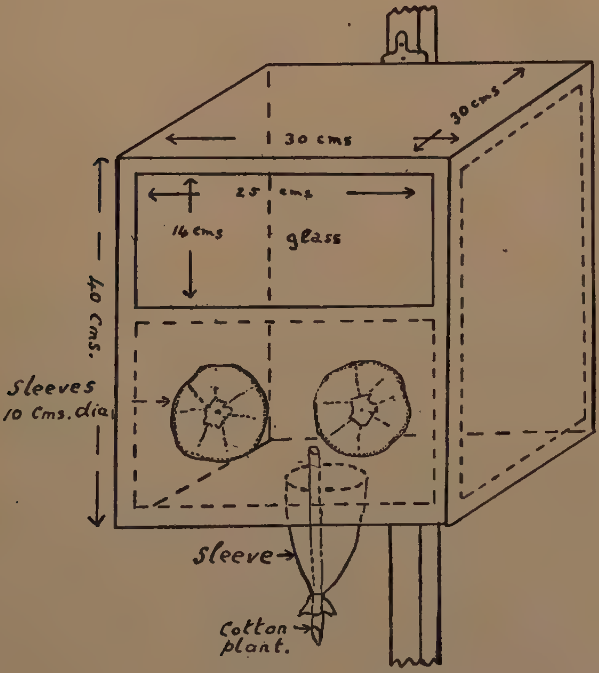

Fig. 1.—Diagram of cage used.

The sweeps were taken by the same operator throughout the season, and as far as possible at the same time of day and against the prevailing wind.

(b) *Cages.*—Owing to the abnormal growth of cotton in the Sudan whenever the plants are grown under insect-proof cages due to the difference of humidity and temperature inside them, the following technique was used in all experimental work on breeding. The cotton was sown on ridges following normal field practice and left uncovered, but frequent inspections were made to destroy any Jassid nymphs that were found. Seven days before the beginning of any tests the plants were carefully freed from any adults or nymphs and covered with a cage which is described above. Before experiments started, each cage was removed and the plants examined to destroy any nymphs which might have hatched from eggs that had been present.

The cages (fig. 1) were constructed of a light wooden framework 30 cm. square and 40 cm. high. The whole was covered with organdie muslin except for a pane of glass 25 cm. × 14 cm. let into the front.

To facilitate handling of the plants and releases of Jassids into the cages, two sleeves were sown into the muslin front below the glass window. The base of the cage was of muslin, in which a large muslin sleeve was sewn to tie firmly round the stem of the cotton plant sufficiently high for the apex of the plant to reach the top of the cage, and so that the cage could be raised to allow for the growth of the plant.

The Seasonal Incidence of Jassids in the Gezira.

Like a number of other insects in the Sudan, Jassids increase at a rapid rate with the arrival of the rains and during the hot weather following the rains, after which

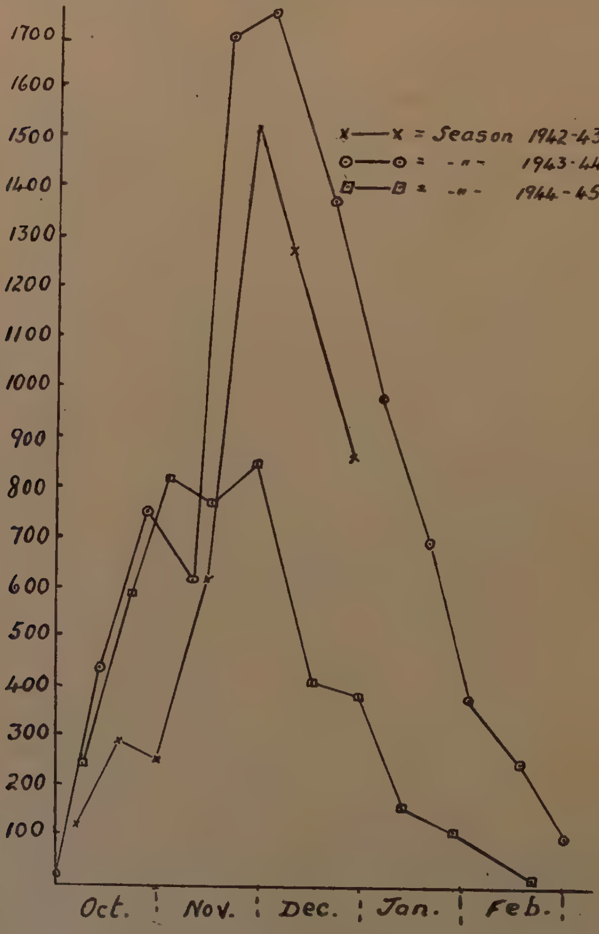

Fig. 2.—Turabi observation plot. Numbers of adult Jassids.

their numbers fall with the onset of the cool, dry conditions of the winter. The adults move to the newly-sown cotton during August almost as soon as the seedlings appear above ground. At first the infestation is small, but the population increases rapidly and reaches a peak about the end of November, after which it decreases fairly rapidly until by the end of February very few Jassids are left on the cotton.

From January onwards Jassids are found in fairly large numbers at first on egg-plants growing on native vegetable patches and then, as the dead season progresses, in smaller numbers on weeds and plants in gardens and on banks where very slow breeding takes place until they disperse to the newly sown cotton in August.

Until the canalisation of the Gezira was extended northwards, symptoms, since ascribed to Jassids, had not been clearly shown. Research was therefore directed towards ascertaining whether the number of Jassids was greater in the north than in the central districts or whether the drier conditions in the former intensified the symptoms.

Sweeping has been carried out during the past three seasons at fortnightly intervals on the Plant Physiologist's Observation Plots at the Gezira Research Farm (central Gezira) and at Turabi (northern Gezira) on all subplots of Massey's selected Domains Sakel.

The damage on these plots was similar to that observed on the neighbouring cotton so that the incidence of Jassids on these plots can be taken as indicative of that in each area.

Figures 2 and 3 show the numbers of adult Jassids taken each fortnight at the two stations for the three seasons 1942/43, 1943/44 and 1944/45. The figures represent 96 forward and backward sweeps taken over 66 metres length.

It will be seen that in all these seasons the numbers of Jassids have been much larger at Turabi (northern area) than at the Gezira Research Farm (central area), but wide differences between the seasons occurred, that were related to the amount of damage observed.

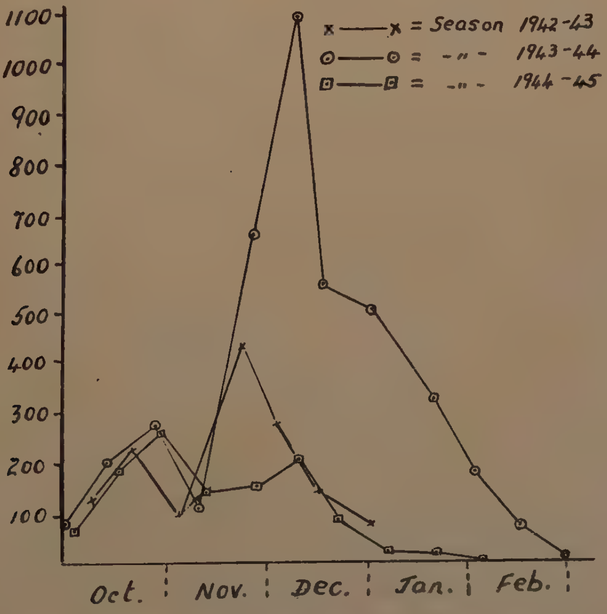

Fig. 3.—Gezira Research Farm observation plot. Numbers of adult Jassids.

The season 1943/44 was a bad Jassid year and scorch was very heavy. Taking the Jassid population throughout the season as 100, that at the Gezira Research Farm was only 46.

In the season 1942/43, the population at Turabi was 54 and at the Gezira Research Farm 15, while in the season 1944/45 at Turabi it was 45 and at the Gezira Research Farm 13.

The following notes, made by the writer during these seasons, show that the numbers are relative to the amount of scorch observed.

Season 1942/43. At Turabi scorch was light up to the end of November, but rapidly increased and was finally fairly heavy. At the Gezira Research Farm scorch was light throughout the season.

Season 1943/44. At Turabi scorch was very severe; at the Gezira Research Farm it was heavy.

Season 1944/45. At Turabi scorch appeared early but was less intense than in the previous two seasons; at the Gezira Research Farm scorch was negligible throughout. In this season the damage promised to be heavy in the northern area, but the early arrival of the cooler weather and dry north winds probably acted as a check to the increase.

The Plant Physiologist kindly supplied figure 4 showing the amount of scorch on a 0–10 scale of damage at Turabi and Gezira Research Farm for the season 1943/44. The graph represents the mean amount of scorch on the main stem leaves at each node. The far heavier damage in the north than in the central area is clearly depicted.

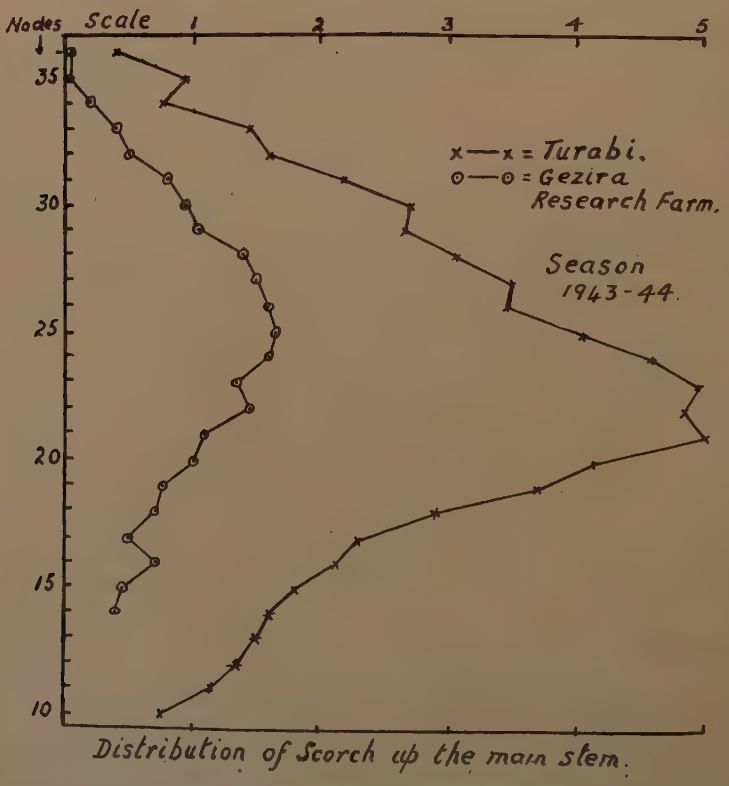

Fig. 4.—Gezira observation plots. Amount of Jassid scorch on 0–10 scale of damage.

During the season 1944/45 sweeping was undertaken on several Gezira blocks distributed through the scheme with the following results :—

TABLE III.

Adult Jassids from 216 forward and backward sweeps.

	November	December	January	Total
Turabi (northern block)	1,927	1,024	475	3,426
Tayiba (east central block)	66	139	74	279
Abdel Galil (central block)	347	414	175	936
Gezira Research Farm (east central block) ...	301	99	74	474
Wad el Bur (western block)	509	565	448	1,522
Ghubshan (southern block)	341	329	89	759

The incidence of Jassids is much greater in the northern and western blocks than in the central and southern blocks, but at present no theories can be put forward to account for this phenomenon.

Parasitism of the adults by a species of *Stylops* has been noted on the Gezira Research Farm to a fair extent but does not appear to be the main factor in control. Parasitism is slight in the north but many more observations are necessary before one can say with certainty what influence it exerts.

Table IV gives the sum of the Jassid population indices on nitrogen-fertilised and control subplots and on two spacings of 80 cms. between rows and 30 cms. and 60 cms. within the rows of Massey Domains Sakel from seven fortnightly sweepings between October and January.

TABLE IV.

Totals from 96 forward and backward sweeps each fortnight.

Spacing		Gezira Research Farm			Turabi		
		Season			Season		
		42/43	43/44	44/45	42/43	43/44	44/45
30 cm.	Control	301	1,076	279	747	2,029	973
30 ,,	Nitrogen	417	952	382	1,567	2,424	1,148
60 ,,	Control	298	1,126	266	1,275	2,131	889
60 ,,	Nitrogen	365	990	226	1,239	2,357	1,062

Statistical analysis of the figures has shown that Jassid population is significantly higher in most years at Turabi on the more luxuriant cotton fertilised at rate of 600 lbs. sulphate of ammonia per feddan*.

In the northern blocks of the Gezira, Domains Sakel cotton is the variety grown, while in the central and southern blocks X1730 Sakel cotton is grown. These two varieties, grown in subplots of a five-feddan plot on the Gezira Research Farm in the three seasons 1942/43, 1943/44 and 1944/45, were swept. To test whether varietal differences affect the population indices, twelve backward and forward sweeps were made on each subplot every fortnight and it will be seen from the results in Table V that in two years, out of the three, Sakel was slightly more heavily infested than X1730A, but there is no statistical difference.

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*1 Feddan equals practically 1 acre.

TABLE V.

Mean Date of Sweeping	1942/43		1943/44		1944/45	
	Sakel	X1730A	Sakel	X1730A	Sakel	X1730A
12 October	162	120	16	22	24	34
25 ,, ...	102	85	96	144	236	240
8 November	147	161	348	301	490	464
22 ,, ...	510	402	439	478	551	451
4 December	353	231	514	868	457	438
18 ,, ...	154	143	936	1,030	195	136
1 January	105	109	591	663	250	241
14 ,, ...	43	40	519	559	—	—
2 February	—	—	335	377	—	—
16 ,, ...	—	—	150	174	—	—
	1,576	1,291	3,944	4,616	2,203	2,004

Experiments on Control of Jassids.

References in the literature, mainly American, on the control of Jassids on an experimental scale have shown that copper sprays have given fairly satisfactory results. It was therefore decided to test out the effect of spraying with Bordeaux mixture. The spray was made as follows :—6 lb. lime putty was dissolved in 32 gallons of water, to which was added 8 gallons of a 1 per cent. solution of copper sulphate, giving a 4–6–40 spray.

In the season 1941/42 a five-feddan plot of Domains Sakel on the Gezira Research Farm was laid out with twelve subplots, six treated and six untreated, of which only the central third of each subplot was picked. Each of the treated subplots was sprayed three times at fortnightly intervals at end of October, mid-November, and end of November, knapsack pressure sprayers being used. Owing to the age of the machines, pressures of over 30 lb. per square inch could not be obtained. The quantity of spray applied per feddan was between 250–300 gallons.

The yields are given in Table VI.

TABLE VI.

Yields in Kantars per feddan.
(1 kantar=315 lbs. seed cotton).
Gezira Research Farm. Season 1941/42.

Untreated	6·27		
Sprayed 3 times ...	6·98	+·71	=11 per cent.

In cooperation with the Chief Plant Physiologist records of certain physiological data were made when carrying out the foregoing experiment.

(*a*) *Plant sampling.*

From each subplot 20 plants were taken at random, *i.e.* 120 plants per treatment. The samples, collected on January 14th, gave the following dry weights in grams.

	Healthy green leaves	Jassid damaged leaves	Total green leaves	Per cent.	Stem	Flower buds	Green bolls	Grand total
Sprayed ...	14·2	2·9	17·1	17	32·2	0·1	44·0	93·4
Unsprayed ...	6·0	11·5	17·5	66	25·9	0·1	41·8	85·3

The number of flower buds in the sprayed samples was 10·5 as against 4·1 in the unsprayed whilst the corresponding figures for green bolls were 2,286 and 2,046.

The dry weights of healthy and Jassid damaged leaves from the sprayed and unsprayed samples reflect the difference in greenness, which was very striking in the field at the time of sampling.

Controlling Jassids caused a slight increase in weight of stem and bolls and an appreciable increase (11 per cent.) in numbers of fruit. Jassids unless controlled evidently lead to premature cessation of main stem and fruit development and hasten the onset of " internal starvation ".

(*b*) *Weight of Seed-Cotton per Boll.*

Samples of healthy bolls were taken ahead of pickings 2–4, and gave the following weights of seed-cotton per boll (grams) :—

	Picking 2 January 28	Picking 3 February 12	Picking 4 February 25
Sprayed ...	3·03	2·82	2·48
Unsprayed ...	2·90	2·69	2·35

At each picking the sprayed plants produced heavier bolls.

(*c*) *Plant Diagrams.*

The distribution of the crop up the main stem was mapped for 8 plants per sub-plot on January 12–13th.

The average numbers of fruits *produced* per main stem node are given below :—

Main Stem Zone	Sprayed	Unsprayed	Difference
Nodes 31–35 ...	1·19	1·00	+·19
„ 26–30 ...	1·33	1·31	+·02
„ 21–25 ...	1·97	1·83	+·14
„ 16–20 ...	2·64	2·42	+·22
„ 11–15 ...	2·19	2·16	+·03
„ 6–10 ...	1·46	1·35	+·11
Total ...	10·78	10·07	+·71

There is extra fruit production with spraying but the data show no trend from the bottom to the top of the plant.

The subsequent fate of these fruits shows more pronounced treatment differences. The proportion of the fruit, at each node, shed before maturity are given below :—

Main Stem Zone	Sprayed	Unsprayed	Difference
	Per cent.	Per cent.	Per cent.
Nodes 31–35 ...	37·5	50·0	−12·5
„ 26–30 ...	45·1	47·6	− 2·5
„ 21–25 ...	32·8	44·9	−12·1
„ 16–20 ...	40·0	38·1	+ 1·9
„ 11–15 ...	40·3	32·7	+ 7·6
„ 6–10 ...	38·1	42·3	− 4·2

Spraying slightly reduces the shedding of fruits in the upper part of the plants, *i.e.* those late formed. Lower down there is *increased* shedding by spraying that, it is suggested, may arise through the act of spraying.

The same experiment was repeated in the 1942/43 season at the Gezira Research Farm and at Turabi except that the belts cut out round each sub-plot was reduced to four metres. The results are given in Table VII and again show a yield increase in favour of the sprayed subplots.

The increase on the G.R. Farm was again 11 per cent. and at Turabi it was 25 per cent.

In the 1943/44 and 1944/45 seasons, in a similar experiment, comparisons were also made between once sprayed at each of the dates against the normal thrice sprayed. Results are given in Table VII.

TABLE VII.

Results of the three year's spraying Experiments.
The Yields are given in Kantars per feddan (1 kantar=315 lbs. seed cotton).

Season		Gezira Research Farm	Turabi
1942/43	Untreated	7·05	4·76
	Sprayed 3 times ...	7·82+·77=11 per cent.	5·93+1·17, or 25 per cent.
	Standard Error ...	±0·186	±·042
	Critical Difference ...	·674 k.p.f.	·188 k.p.f.
1943/44	Untreated	5·57 } 5·585	4·20 } 4·24
	Untreated	5·60 }	4·28 }
	Sprayed end Oct. only	6·22+·65=11 per cent.	4·97+ ·73=17 per cent.
	" mid Nov. only	6·18+·60=11 "	5·48+1·24=29 "
	" end Nov. only	5·89+·31=6 "	5·31+1·07=25 "
	" three times	6·63+1·05=19 "	5·87+1·63=38 "
	Standard Error ...	±·180	±·180
	Critical Difference ...	·523 k.p.f.	·508 k.p.f.
1944/45	Untreated	6·80 } 6·675	6·83 } 6·84
	Untreated	6·55 }	6·85 }
	Sprayed end Oct. only	6·25—·43	7·65+ ·81=⊕12 per cent.
	" mid Nov. only	6·51—·17	7·50+ ·66=⊕10 "
	" end Nov. only	7·10+·42	7·50+ ·66=⊕10 "
	" three times	6·37—·31	8·18+1·34=20 "
	Standard Error ...	±·37	±·185
	Critical Difference ...	—	·522 k.p.f.

Turabi results.

In all three seasons spraying, whether single or triple applications, significantly increased the yield. The three sprayings combined was significantly better than single sprayings. Comparing the single sprayings at the separate dates, differences just failed to reach significance.

The sprayed plots were much greener and showed far less hopperburn than the untreated plots. An increase in yield of over a kantar was obtained each season from the triple spraying, the largest being 1·83 kantars per feddan in 1943/44, and increases in yield of half to over one kantar from the once sprayed plots.

G.R. Farm results.

The increase was not so great as at Turabi but significance was obtained in all seasons except 1944/45 for the triple sprayings and for the single sprayings at end of October and mid-November but not for the end of November treatment.

In 1944/45 Jassid incidence was low and very little hopperburn was visible even on the untreated subplots. The big differences in the yield of the two unsprayed treatments negatived any likely results. The increase in yield of the end of November spraying may be due to the full cover and larger amount of liquid sprayed per feddan when applied by knapsack sprayers, as against the smaller amount applied at the two earlier applications with a horse drawn barrel sprayer mounted on a cart.

This barrel sprayer was fitted with seven double nozzles to spray seven rows of cotton but it only delivered about 80 gallons per feddan as against the usual rate of 250 gallons per feddan with the knapsack sprayers required to obtain a good cover.

It is possible that a heavy cover with Bordeaux mixture is necessary to obtain a good protection from Jassid damage and that light applications are ineffective. The lower yields of the early sprayed plots was not due to damage done by the machine as it had to travel through all subplots in order to turn at the head lands.

Figure 5 gives the number of adult Jassids and nymphs taken at fortnightly intervals from each subplot of the spraying experiment at Turabi during the season 1944/45.

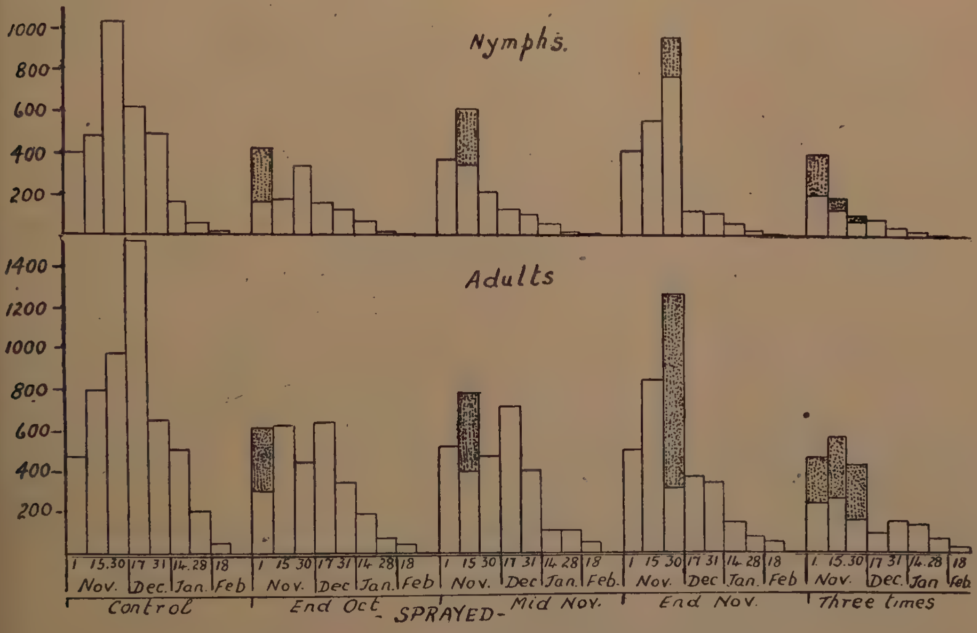

Fig. 5.—Spraying experiment at Turabi—Season 1944/45.

The shaded area represents the reduction in numbers two days after the application of the spray.

The increases in yield obtained by controlling Jassids suggest that it would be a commercial proposition but the difficulty experienced in the preparation of Bordeaux mixture would rule it out as a method to be used on an extensive scale. Trials have therefore been started with DDT and Gammexane and these give promise of better results.

For the seasons 1943/44 and 1944/45, the late Dr. F. Crowther kindly undertook to make an estimation of the amount of Jassid scorch on the Turabi spraying experiment. The counts were made during the first week of February and figs. 6 and 7 show the results.

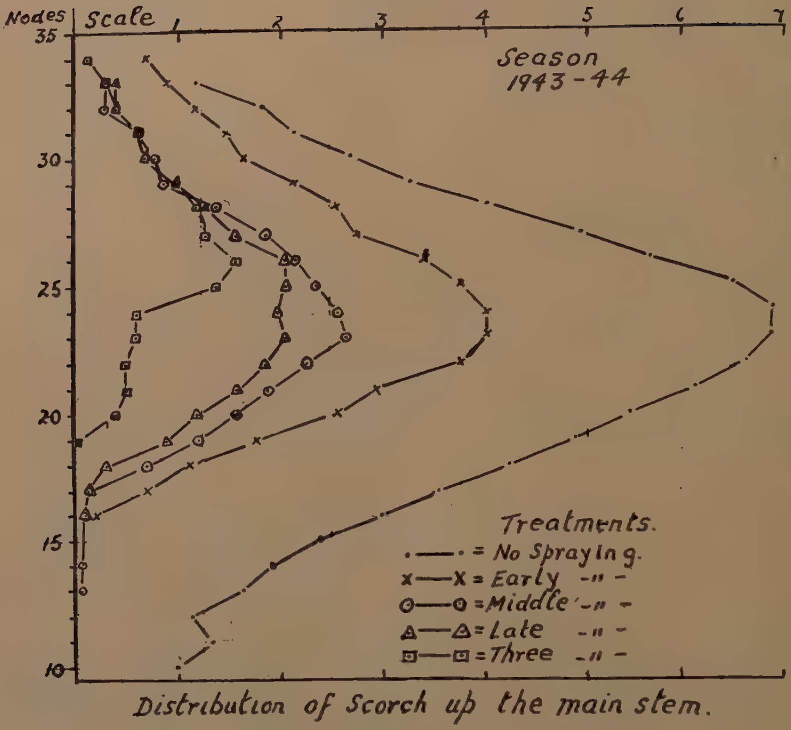

Fig. 6.—Turabi. Leaf scorch severity on 0–10 scale.

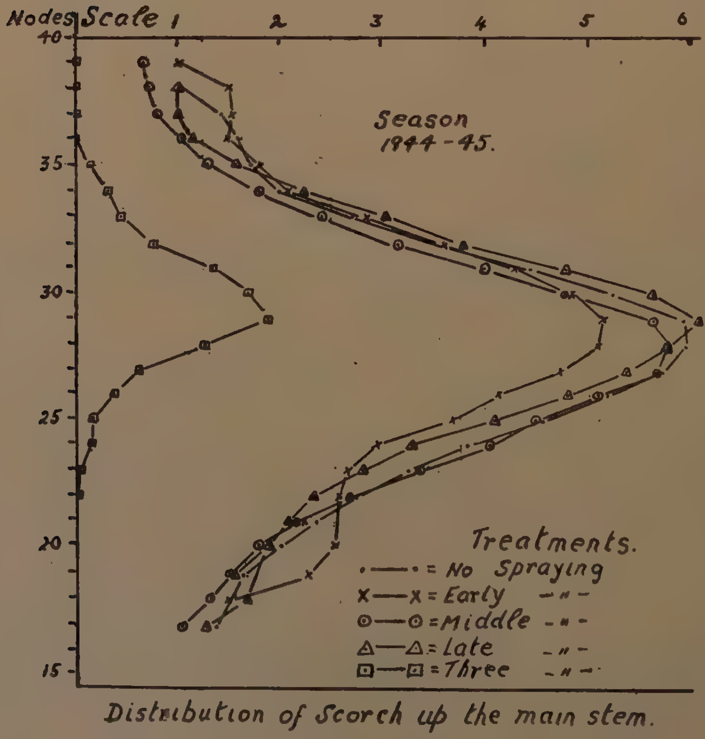

Fig. 7.—Turabi. Leaf scorch severity on 0–10 scale.

Search for Resistance in Sakel Varieties.

Dr. Knight, Senior Economic Geneticist, gave the writer a number of varieties to test for Jassid resistance. Tests were made under cages and in the field in replicated lines with the following varieties: the original parents Tanguis, and Sakel NT/2/39 and their F1 cross and their F2 to F7 backcrossed with Sakel NT/2/39; Hairy leafed Sakel; Domains Sakel; and X1730A Sakel.

Tanguis is a hairy-leafed cotton which has shown considerably less hopperburn in the field but has not the lint quality of a Sakel.

Test under cages.

These varieties were sown in a plot on ridges under normal field conditions and kept as free as possible from Jassids until such time as the plants were caged. The cages used have been described earlier. Six plants of each variety were caged and infested with 5 pairs of Jassids collected from the field. These insects were left on for 12 days when the cages were removed and the number of adults still alive were released. Any nymphs that had hatched were counted and removed before the cages were replaced. At every five-day period the cages were removed and all nymphs counted and removed until no further nymphs emerged.

Table VIII gives the total nymphs that hatched on each variety.

TABLE VIII.

Each variety	Total nymphs	Each variety	Total nymphs
Tanguis	545	F.6 backcross	569
NT/2/39	450	F.7 ,,	397
F1 cross	354	Hairy X1530	514
F2 backcross	541	Domains Sakel	457
F3 ,,	397	X1730A	521
F4 ,,	665		
F5 ,,	464		

It will be seen that not one of the varieties was more than slightly resistant.

Field Tests.

Three rows of each of the varieties were sown at the Gezira Research Farm and Turabi in randomized blocks with the control NT/2/39 between every 2nd line. Nymphs were counted on ten mainstem leaves from two plant holes in each line at fortnightly intervals throughout the highest infestation period from October to December. The results are given in Table IX.

TABLE IX.

	Gezira Research Farm		Turabi	
	Total nymphs	Nymphs per plant holes	Total nymphs	Nymphs per plant holes
NT/2/39	608	19·0	514	85·6
Tanguis	190	31·7	330	55·0
F.1 cross	196	32·7	478	79·7
F.2 backcross	179	29·8	498	83·0
F.3 ,,	165	27·5	428	71·3
F.4 ,,	174	29·0	654	109·0
F.5 ,,	178	29·7	499	83·2
F.6 ,,	201	33·5	478	79·7
H 1530 ,,	122	20·3	531	88·5
X 1730A ,,	123	20·5	499	83·2
Domains Sakel	152	25·3	438	73·0

Again no variety gave a marked reduction in the number of nymphs that hatched although there was less damage apparent on some of the varieties. Tanguis showed less hopperburn than the other varieties, but this may be largely due to the greater number of leaves and more branching habit, or to its greater resistance to the penetration of the stylets of the bugs. But this resistance hardly showed in the first cross and certainly not in the succeeding backcrosses.

Acknowledgements.

The work was carried out at the Gezira Research Farm and at Turabi on the Sudan Plantations Syndicate land. I wish to express my thanks to the members of the Agricultural Research Division, especially to the late Dr. F. Crowther and his staff, for supplying the data for Figures 4, 6 and 7, and for supervising the agricultural work and picking at Turabi; to the Sudanese Entomological Staff, Abdulla Eff. Hamid and Mustafa Eff. El Sawi, and to Abdel Gadir Eff. Farag for the preparation of the spray, and to the Inspectors at Kabel Gidad Block for their hospitality and interest in the work.

Summary.

Empoasca libyca is a major pest of cotton in the northern and western blocks of the Gezira Irrigated Scheme.

Distribution of the species of the genus of *Empoasca* and *Erythroneura lubiae* is given, together with their food plants in the Sudan.

Damage caused by *E. libyca* is noted; an experiment carried out in 1935/36 showed that Jassids were able to cause such symptoms.

The life-cycle shows that the egg and nymphal stages are of short duration (8 days each) and followed by a long adult life (as long as 2 months) in which the female lays her eggs over a long period.

A new type of cage was used and is figured.

The effect of applications of Bordeaux mixture in reducing the Jassid population and amount of scorch is given as well as physiological data relating to the influence of spraying on different parts of the Jassid-infested cotton plants. It is shown that sprayed plants produced an appreciable increase in the number of green bolls formed and an increase of 11 per cent. in the weight of seed cotton per boll harvested. It is pointed out, however, that Bordeaux mixture has several disadvantages and other more suitable insecticides are being sought.

No Sakel varieties as yet have shown resistance to Jassid attack.

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FINAL SURVEY OF THE WOOD-DESTROYING INSECTS IN PUBLIC BUILDINGS IN SWEDEN.

By Ivar Trägårdh.

E.M.N. 4. 24

Formerly Chief, Entomological Department, R. Swedish Institute of Experimental Forestry.

Short summaries have previously been given (1938, 1940) of the results obtained during investigations on the wood-destroying insects in public buildings in Götaland and Svealand, the southern and middle parts of Sweden. The survey has since been continued in Norrland, the northern part of the country, and a short summary of the results obtained may now be given, together with a general survey of the conditions prevailing in the whole country.

Altogether 13,200 detailed questionnaires were sent out, each one accompanied by a circular letter and a leaflet containing short descriptions of the insects in question and illustrations of the damage caused by them. Of these a little over 5,000 reports were returned, 2,122 of which gave fairly detailed information and many of these were accompanied by samples of wood for identification of the insects that had caused the injury.

TABLE I.

Percentage of all injuries in the three main parts of Sweden.

	Questionnaires	Answers	Percent.	Injured buildings	Uninjured buildings	Percent. injured
Götaland	7,000	2,148	30·0	990	1,158	46·1
Svealand	4,900	1,801	36·7	789	1,012	43·8
Norrland	1,300	1,113	85·6	383	770	31·0

Table I shows that the percentage of damaged buildings is only 31 in Norrland as against 46·1 in Götaland and 43·8 in Svealand. The same insects were reported as in the earlier surveys, viz., *Hylotrupes bajulus*, L., *Callidium violaceum*, L., *Ernobius mollis*, L., *Anobium striatum*, Oliv., *Monochamus sutor*, L., and *Sirex* spp.

The figures in Table I are, however, of only limited value unless they are supplemented by information regarding *which* insects are concerned in the injuries in different parts of the country.

In the previous reports the insects were classified into three distinct groups. It seems advisable to repeat this classification here for the sake of completeness.

Group I.—Passive intruders that are introduced in the larval or pupal stage with the timber but, whilst completing their development, are unable to propagate in houses. The source of the infestation is the forest, and is due to the practice of leaving undecorticated trees or logs in the forest during the summer or the use of timber from dead trees.

This group embraces *Monochamus sutor* and several species of *Sirex* and occurs comparatively rarely.

Group II.—Passive intruders, the larvae of which tunnel under the bark and are generally introduced into the houses at the time of building, if bark is left on the timber. The source of infestation is generally saw-mills and timber yards. This group embraces *Callidium violaceum* and *Ernobius mollis*, both of which may continue to breed in the houses for some time, provided that there is enough uninfested timber with bark left on it.

Group III.—Permanent, active intruders, living in the wood and as a consequence not dependent on the presence of bark. They continue their attacks in the houses generally for many years. This group embraces *Hylotrupes bajulus* and *Anobium striatum*.

In Table II information regarding the last two groups, that are by far the most important, is given. The most striking result is that *Hylotrupes bajulus*, the most important pest, is practically non-existent in Norrland and that *Anobium striatum*, the second worst pest, is also very rare.

TABLE II.

Percentage of Government Buildings of different kinds reported as attacked in Götaland (G), Svealand (S) and Norrland (N).

		Churches	Public buildings	Dwelling houses	Outhouses	All buildings
H. bajulus ...	G.	16·6	28·0	15·2	11·7	15·8
	S.	20·0	32·4	19·8	37·0	26·9
	N.	—	—	2·4	2·5	2·0
A. striatum	G.	72·9	53·0	66·2	72·7	68·1
	S.	55·4	33·5	43·3	55·2	48·9
	N.	15·4	2·3	3·6	—	2·6
E. mollis ...	G.	19·6	39·0	44·7	43·6	38·8
	S.	33·1	32·4	40·2	31·0	35·2
	N.	46·2	47·7	30·3	32·2	33·8
C. violaceum	G.	7·5	22·0	25·9	23·4	21·1
	S.	24·6	55·9	37·8	42·4	38·1
	N.	84·6	68·2	74·5	80·3	76·1

If a comparison is made between the three principal parts of Sweden of the number of attacks of these four insects in relation to all the buildings from which reports, either negative or positive, were returned, one gets a very good idea of their different frequency in these parts. It will be noticed that *Hylotrupes bajulus* is almost twice as common in Svealand as in Götaland, but exceedingly rare in Norrland where it has only been reported from the southernmost part. *Anobium striatum*, on the other hand, decreases gradually northwards and is quite as rare in Norrland as *H. bajulus*. Considering the great economic importance of these two beetles, this result which, by the way, tallies perfectly with what is known about their geographical distribution from other sources, is of very great interest.

Callidium violaceum occurs all over the country, increasing in frequency northwards, while *Ernobius mollis* also occurs all over the country but decreases slightly northwards.

The two earlier surveys presented the striking feature that the counties round the coasts were infected on the average twice as heavily as those in the interior. When discussing this phenomenon (1940), I put forward several suggestions. Granted that the most injurious of these insects (Group III) spread from building to building and from village to village through active means, *viz.*, by using their wings, a greater density of population, which is the same as a shorter distance between the houses, must be considered to favour the spreading of the species to such an extent that the infestation in the densely populated parts becomes more severe. As a matter of fact there is a very good correlation between the density of the population and the frequency of the attacks. The mean in the coast zone is 50·2 inhabitants per square km., in the other zone 25·5, the respective percentages of houses attacked being 49·8 and 25·6. Thus the possibility that there is a causal connection between the density of population and the frequency of attack must not be overlooked.

Finally there is the possible influence of climatic conditions to be considered, although admittedly it is impossible to gauge their effect on insects in houses by using the generally adopted method of constructing climograms. For the same reason the investigation was restricted to the temperature, and more particularly to the mean temperature of the coldest month, February, and the yearly mean temperature. The former is only —1·25° Celsius in the coast zone but —3·08° Celsius in the inland zone, the mean annual temperature being in the coast zone +7·11° Celsius and in the inland zone +5·4° Celsius. Until experimental evidence is available regarding the influence of different temperatures on the viability and development of these insects it is impossible to draw any conclusions from these data.

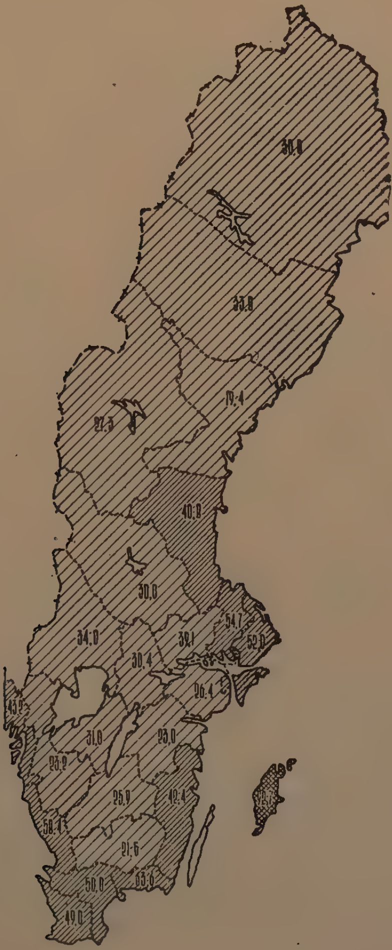

Fig. 1.—Map of Sweden showing the percentage of public buildings attacked by the four most important species of wood-destroying insects, viz., *Hylotrupes bajulus*, *Anobium striatum*, *Callidium violaceum* and *Ernobius mollis*.

It was impossible to test the influence of atmospheric humidity on account of the insufficient funds allotted to the investigations; but in the meantime Danish and German authors have investigated the influence of these two factors on *Hylotrupes bajulus*. It may now be considered an established fact that an air humidity of 90–95 per cent. is favourable to the propagation of these insects in so far as the mortality of the eggs is lowered and the larval development accelerated.

The map, fig. 1, shows the percentage of government buildings reported as attacked by wood-destroying insects in the different counties of Sweden. As in the previous surveys three degrees of infestation were recognized, viz., below 40 per cent., 40–75 per cent. and above 75 per cent. The heavier infestation of the coast region extends only to Gästrikland, north of Uppland, a fact which is probably accounted for by the absence further north of group III. The only exceptions from the rule that the coast region is more severely infested than the inland zone are Linköpings and Södermanlands counties. But a study of the configuration of these counties shows that, although formally belonging to the coast zone, their coast line is actually very short, the main part belonging to the interior.

TABLE III.

The frequency of *Hylotrupes bajulus* and *Anobium striatum* in public buildings in Götaland and Svealand.

	H. bajulus	*A. striatum*	Both species
GÖTALAND			
Göteborgs and Bohuslän ...	2·8	30·2	31·7
Hallands län	7·3	49·1	49·1
Malmöhus län	10·7	40·5	46·4
Kristianstads län	7·8	40·9	43·4
Blekinge län	19·2	42·3	52·6
Kalmar län	9·1	29·3	33·5
Älvsborgs län	3·3	9·9	9·9
Skaraborgs län	0·5	13·5	13·9
Jönköpings län	2·1	9·1	9·6
Kronobergs län	1·1	12·1	13·2
Östergötlands län	3·1	15·4	16·4
SVEALAND			
Stockholms län	14·6	18·4	28·5
Uppsala län	18·8	19·6	32·4
Södermanlands län	2·8	11·0	12·5
Västmanlands län	—	15·2	15·2
Örebro län	4·3	24·3	24·3
Kopparbergs län	—	4·2	4·2
Värmlands län	16·1	18·5	20·3
Gotlands län	27·4	79·0	87·1

On the map the injuries caused by groups II and III are included, but in order to estimate more exactly the economic losses caused it is necessary to consider the two most injurious species belonging to group III alone, *viz.*, *Hylotrupes bajulus* and *Anobium striatum*. Table III gives the data obtained from this investigation. The figures give the attacks of each species and of both species together, since they often occur together, in percentages of all the buildings from which reports were received. In the writer's opinion the figures are in some instances quite alarming, as for instance 52·6 per cent. in Blekinge and above 30 per cent. in several other counties.

The percentage of churches injured is very low in Norrland and this seems to be explained by the fact that *Hylotrupes bajulus* and *Anobium striatum*, that in other parts of the country are so prevalent in churches, are exceedingly rare in Norrland.

In the previous surveys an attempt was made to find out the frequency of these last two species in the different parts of buildings. In both Götaland and Svealand the injury was concentrated in the roof-timbers. In Norrland, where these two species are practically missing and *Callidium violaceum* and *Ernobius mollis* are the dominating species, the roof-timbers are, nevertheless, even more attacked than other parts of the buildings, obviously because the roof-timbers have bark left under which these species find a suitable habitat.

The fact that both groups II and III dominate in the roof-timbers makes it interesting to see whether the frequency of attack is influenced by different roofing materials. These may act in several ways, either through the colour of the material or its density or through its insolating power. The timber under dark roofs is apt to grow hot through insolation, while roofs of a more porous material may facilitate ventilation and thus tend to keep the temperature down.

The investigations of Götaland elicited the fact (1940, p. 291) that houses with iron roofs were most attacked (21·3 per cent.). Also in Svealand similar houses were more commonly attacked than houses with other types of roof (28 per cent.), but houses covered by shingle were even more heavily attacked (34·2 per cent.) (1940, p. 292). The results obtained over the whole of Svealand were shown in *l.c.* fig. 3. The most remarkable feature is the very pronounced increase in the injury in houses with tile-roofs and the less marked increase in houses with iron roofs. Whether the roofing-material has any effect on the frequency of the attacks, and if so how, is, of course, very complicated and can only be solved by experimental researches. The statistical data so far collected in different countries do not tally very well. In Germany, for instance, where an immense amount of data has been collected, houses with slate roofs were most attacked, houses with iron roofs being least attacked.

There is, after all, nothing remarkable in this discrepancy because it is obvious that it is not the roofing material in itself which is the deciding factor, but the way it reacts under the prevailing climatic factors to produce a temperature and degree of atmospheric humidity favourable to the insects in question. It may be that in the continental climate of Germany the temperature rises so highly under the iron roofs that it becomes lethal to the insects, whereas in Sweden with its colder climate the same roofs are favourable to the insects.

In the first survey (1938, p. 62) an attempt was made to investigate the influence of the age of the houses on the frequency of attacks in Götaland. Altogether 637 reports, or 70·5 per cent., gave information regarding this point. The houses were divided into ten age-groups of ten years each, and the number of houses of each class were plotted. It was found that the percentage of attack rose with increasing age up to 30 years, after which it remained fairly constant. It would perhaps be premature to interpret this as indicating that the insects prefer old timber to new. Rather it expresses the fact that with increasing age the chances of infestation increase, until the upper limit of infestation is reached after 30 years, when no further increase is possible, because all possible suitable localities have been exploited.

The data from Svealand (1940, p. 293) tally fairly well with those from Götaland. In this investigation *Anobium striatum* was more frequent than *Hylotrupes bajulus* and the percentage of attacked houses rose with increasing age up to 40–70 years, when no further increase occurred. These results agree with those obtained from Germany where it was found that the younger the houses the less were they attacked and that after 50–60 years the percentage of attacks was fairly constant.

In Norrland, as mentioned above, only *Callidium violaceum* and *Ernobius mollis*, that are not permanent pests, have to be dealt with, the duration of their attack being

TABLE IV.

Influence of the age of houses on the attack in Norrland.

Years	1–10	11–20	21–30	31–40	41–50	51–60	61–70	>70
Number of houses ...	237	132	125	115	91	39	25	140
„ „ attacks ...	9	12	18	9	5	2	2	0
Percentage	3·8	8·1	17·7	7·8	5·5	5·1	8·0	0

limited by the supply of bark under which they live. This difference affects the frequency of their attacks in houses of different ages (Table IV). The percentage rises rapidly from 3·8 to 9·1 and 14·4 during the first 30 years, but goes down afterwards, obviously because the supply of bark has become exhausted. The rise in houses older than 60 years is only apparent, because it is based on only two attacks.

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ON THE EFFECT OF DDT-TREATED SURFACES ON ADULTS OF *ANOPHELES GAMBIAE* AND *A. FUNESTUS*.*

By D. G. MacInnes.

Introduction.

In April 1945 preliminary air-spray trials were instituted at Dar-es-Salaam as a larval control measure. The spray mixture used was a 4·0 per cent. (pure) DDT weight/vol. solution, in equal parts of Auto-Gas-Oil and Furnace Oil, and was applied at the rate of 1·5 quarts per acre, giving approximately 17 mgms. (pure) DDT per sq. metre. In addition to the larval effects, which are being described in detail elsewhere by Wilson & Robertson, it was found that after application of the air-spray, adult vector catches from the sprayed area were not immediately reduced, although the anticipated seasonal rise did not occur.

Further air-spray trials were carried out at Kisumu from July to October 1945 by the same workers when the spray concentration of DDT (pure) was increased to 5·0 per cent., and dosage increased to 2·25 quarts per acre, or approximately 32 mgms. DDT (pure) per sq. metre. In this trial the adult vector population in the air-spray controlled catching stations was reduced by 98 per cent. within three days of the first application of the spray.

Laboratory tests were carried out at the Taveta research station of 92 (EA) Malaria Field Laboratory, E.A.A.M.C., between January and April 1946, in order to investigate the cause of this remarkable effect upon the adult mosquito population resulting from larvicidal air-spraying with DDT. Certain questions arising from these experiments were also examined, and the results of each are tabulated below.

The Effect on Adult Anopheles of DDT Films on Water.

Procedure.—Adult ♀♀ of both vector species were caught, and approximately equal numbers placed in two cages each of 1 cu. ft. capacity. Each cage contained a bowl of water; in the one case the water was treated with a DDT solution, whilst in the other it was left untreated as a control. The mosquitos were caught during the morning, sorted and caged during the afternoon, and the final observations made during the following morning, after approximately 18 hours in the cages.

The different effects of DDT solutions in power kerosene and in the air-spray mixture were examined.

Experiment 1.

Mixed ♀♀ of *Anopheles gambiae*, and *A. funestus*, Giles, of all stages of ovary development were used. A 5·0 per cent. solution of DDT in power kerosene was applied to the water of one bowl, at the rate of approximately 200 mgms. DDT (pure) per sq. yard. It was found in this experiment, and in all subsequent trials with kerosene solutions, that the solvent evaporated within 20–30 minutes, leaving a film of DDT crystals floating on the water surface (in cold weather the evaporation period extended up to 2 hours). The crystals so formed were in rosettes and needles that remained floating for more than 24 hours, even after extensive stirring.

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The results of this test were as follows :—

TABLE I.

The effect of a water-surface film of 5·0 per cent. DDT in kerosene applied at the rate of 200 mgms. DDT per sq. yd., on adult vectors of all stages of ovary development.

Control Cage						DDT Cage					
Total No. of Vectors	Laid		Did not lay		Mort. %	Total No. of Vectors	Laid		Did not lay		Mort. %
	Alive	Dead	Alive	Dead			Alive	Dead	Alive	Dead	
40	0	0	38	2	5	40	0	1	4	35	90
30	0	3	20	7	33	30	0	2	5	23	83
76	1	4	34	37	54	64	0	2	31	31	51
71	0	11	25	35	65	69	0	5	22	42	68
51	0	10	15	26	70	24	0	2	1	21	96
13	0	0	11	2	15	14	0	0	8	6	75
14	0	0	9	5	36	22	0	0	5	17	77
56	0	7	37	12	34	61	1	7	36	18	41
23	0	0	20	3	13	21	0	0	10	11	52
47	0	4	37	6	21	45	0	3	19	23	58
48	0	3	41	4	14	44	0	1	33	10	25
50	0	3	39	8	22	52	0	1	34	17	35
519	1	45	326	147	37	486	1	24	208	254	57
	0·2%	8·7%	62·8%	28·3%			0·2%	5·0%	42·7%	52·1%	

In the Control series 9 per cent. laid, of which 98 per cent. died.
In the DDT series 5 per cent. laid, of which 96 per cent. died.

Experiment 2.

The same procedure and dosage was adopted, but in this test only ♀♀ of ovary stages IV–V were used.

TABLE II.

The effect of a water-surface film of 5·0 per cent. DDT in kerosene applied at the rate of 200 mgms. DDT per sq. yd., on adult vectors of ovary stages IV–V only.

Control Cage						DDT Cage					
Total No. of Vectors	Laid		Did not lay		Mort. %	Total No. of Vectors	Laid		Did not lay		Mort. %
	Alive	Dead	Alive	Dead			Alive	Dead	Alive	Dead	
9	2	2	2	3	55	8	0	5	0	3	100
5	0	1	1	3	80	10	1	1	0	8	90
6	2	0	2	2	33	9	0	1	2	6	78
15	1	2	3	9	73	16	2	1	7	6	44
8	1	5	2	0	62	14	1	2	5	6	57
12	0	2	5	5	58	13	0	2	0	11	100
18	4	5	3	6	61	20	1	3	4	12	75
3	0	0	0	3	100	3	0	1	1	1	67
76	10	17	18	31	63	93	5	16	19	53	74
	13%	22%	24%	41%			5%	17%	21%	57%	

In the Control series 36 per cent. laid, of which 63 per cent. died.
In the DDT series 22 per cent. laid, of which 76 per cent. died.

Experiment 3.

The procedure was similar to that of the first two trials, but in this test the two vector species were tested separately. Only ♀♀ of ovary stages IV–V were used.

The bowls were treated with the same solution of DDT in power kerosene, applied at the rate of approximately 40 mgms. DDT (pure) per sq. yard.

TABLE III.

The effect of a water-surface film of 5·0 per cent. DDT in kerosene, applied at the rate of 40 mgms. DDT per sq. yd., on ♀♀ of *A. gambiae* and *A. funestus* in ovary stages IV–V.

(a) *A. gambiae.*

Control Cage						DDT Cage					
Total No. of Vectors	Laid		Did not lay		Mort. %	Total No. of Vectors	Laid		Did not lay		Mort. %
	Alive	Dead	Alive	Dead			Alive	Dead	Alive	Dead	
22	7	3	12	0	14	22	4	4	9	5	41
23	11	3	7	2	22	24	1	5	3	15	83
15	6	3	2	4	47	15	0	9	2	4	87
17	7	1	5	4	29	15	0	1	3	11	80
13	3	2	7	1	23	14	0	4	8	2	43
18	4	5	5	4	50	19	0	3	2	14	89
108	38	17	38	15	30	109	5	26	27	51	76
	35%	16%	35%	14%			4·5%	24%	25%	46·5%	

(b) *A. funestus.*

13	7	3	0	3	46	14	1	2	2	9	79
28	11	2	7	8	36	27	1	6	2	18	86
8	1	1	5	1	25	12	0	3	2	7	83
8	0	2	3	3	63	7	0	0	3	4	57
22	3	6	2	11	77	22	1	9	1	11	91
26	5	7	3	11	69	23	0	3	0	20	100
105	27	21	20	37	55	105	3	23	10	69	88
	26%	20%	19%	35%			3%	22%	9%	66%	

In the Control series of *A. gambiae*, 51 per cent. laid, of which 30 per cent. died.
In the DDT series of *A. gambiae*, 28 per cent. laid, of which 84 per cent. died.
In the Control series of *A. funestus*, 46 per cent. laid, of which 44 per cent. died.
In the DDT series of *A. funestus*, 25 per cent. laid, of which 88 per cent. died.

Experiment 4.

For this experiment the effect of the solution used in the air-spraying was tested, and the dosage was designed to be approximately the same as that of the Kisumu air-spray trials. The results were as follows :—

TABLE IV.

The effect of a water-surface film of 5·0 per cent. DDT in equal parts of AGO and Furnace oil, applied at the rate of 35 mgms. DDT per sq. yd., on ♀♀ of *A. gambiae* and *A. funestus* in ovary stages IV–V.

(a) *A. gambiae.*

Control Cage						DDT Cage					
Total No. of Vectors	Laid		Did not lay		Mort. %	Total No. of Vectors	Laid		Did not lay		Mort. %
	Alive	Dead	Alive	Dead			Alive	Dead	Alive	Dead	
14	6	1	6	1	14	13	0	0	4	9	69
7	2	1	2	2	43	9	1	1	0	7	90
16	0	0	15	1	6	16	0	1	9	6	44
14	2	2	9	1	21	13	0	0	6	7	54
12	3	1	6	2	25	13	0	1	4	8	69
18	8	5	3	2	39	20	1	3	2	14	85
81	21	10	41	9	23	84	2	6	25	51	68
	26%	12%	51%	11%			2%	7%	30%	61%	

(b) *A. funestus.*

8	3	3	2	0	38	7	0	0	0	7	100
7	2	2	2	1	43	8	0	0	0	8	100
22	2	7	10	3	45	22	0	0	3	19	86
9	1	6	2	0	66	16	0	1	4	11	75
9	0	4	3	2	66	10	0	0	1	9	90
55	8	22	19	6	51	63	0	1	8	54	87
	14·5%	40%	34·5%	11%			0%	1·6%	12·7%	85·7%	

In the Control series of *A. gambiae*, 38·0 per cent. laid, of which 22 per cent. died.
In the DDT series of *A. gambiae*, 9·5 per cent. laid, of which 75 per cent. died.
In the Control series of *A. funestus*, 54·5 per cent. laid, of which 73 per cent. died.
In the DDT series of *A. funestus*, 1·6 per cent. laid, of which 100 per cent. died.

On the one occasion recorded when *A. funestus* laid on treated water, laying was incomplete, only nine eggs being produced.

In this trial the oil film retained the DDT in solution for more than 24 hours.

Discussion.

The high mortality in the control series throughout these experiments appears to have been due mainly to the prevailing climatic conditions. This is particularly apparent in experiment 2, which was, in fact, the first test to be carried out, when mortality from unfavourable conditions was at its height. Classifying the results into (i) Alive : Dead, and (ii) Control : DDT, and using the statistical test for the independence of these two classifications, the values of X^2 are found to be significant at the ·01 level in all the experiments but No. 2 in which they are not significant even at the ·05 level (Table V).

TABLE V.

Statistical comparison of mortalities in each test.

Experiment No.	Value of X^2	X^2 ·05	X^2 ·01
1	40·7	3·841	6·635
2	2·47	3·841	6·635
3(*a*)	36·8	3·841	6·635
3(*b*)	26·8	3·841	6·635
4(*a*)	32·8	3·841	6·635
4(*b*)	18·6	3·841	6·635

These figures indicate that in these experiments, with the exception of No. 2, a greater number than could have been expected by chance died in the DDT cage than in the Control cage. On the other hand, the variability within the samples in each experiment renders statistical comparison a matter of somewhat doubtful value, and further tests along these lines would be necessary before the exact degree of significance could be determined.

In all experiments a greater number of mosquitos laid in the control cages than in those with treated water, particularly in No. 4, when the DDT was applied in a more persistent oil. This suggests a direct inhibitory action. Statistical comparison of the numbers that laid in each cage again shows a significant difference throughout, except in No. 2, but in view of the low figures in each test, such comparison is open to doubt.

Ratio of Mortality : Period of Exposure on treated Wood Surfaces.

In experiment 1 above, the effect of a water-surface film of DDT on vector adults in all stages of ovary development, was tested. It was anticipated that no significant difference in mortality would be found, except amongst the mosquitos that laid, but since the increased total mortality in the DDT cage proved to be highly significant, it may be assumed that a ♀ mosquito, even in an early stage of the ovary cycle, may be attracted to, and come into contact with, water if it is reasonably accessible, despite the presence of a DDT film.

Hocking & MacInnes (unpublished) found that African vector Anopheline females often rest on the water surface whilst laying, and that a healthy *Anopheles gambiae* is capable of laying the normal complement of eggs in a space of 5 minutes. For this reason a few experiments were carried out in order to determine the period of exposure necessary for an adult mosquito to acquire a lethal dose of DDT.

Procedure.

A wooden box was designed and constructed, consisting of four sides, with a sliding glass top ; the bottom was formed by a small table, the area of which was more than double the basal area of the box. The inside was fitted with four side panels of 3-ply wood, two of which could be withdrawn through slots in the table top, whilst the other two could be withdrawn upwards after sliding the glass top as required. The panels were treated on the inner surface with a solution of DDT in power kerosene, applied at the rate of 200 mgms. per sq. foot. Three panels were placed in position, and the mosquitos inserted through a sleeve let in to the fourth side, the remaining panel being slipped into place immediately after releasing the mosquitos. The insects were thus enclosed in a space with four vertical treated surfaces, and an untreated top and floor. After an appropriate interval of exposure, the panels were withdrawn simultaneously, and the box was moved to the other end of the table to eliminate any possibility of DDT crystals, that might have been scraped off the panels, remaining on the floor. By this means the various periods of exposure could be timed accurately, and at the same time the mosquitos were not subjected to any unnecessary handling.

Owing to a breakdown in the apparatus, no experiments were made until 5 months after the impregnation of the panels.

The following table expresses the results of several tests at each period of exposure, in terms of mortality percentage :—

TABLE VI.

Ratio of Mortality : Period of exposure to DDT surface.

Mins. Exp.	No.	Mortality (from end of exposure period) at :									
		0 hr.	½ hr.	1 hr.	2 hrs.	3 hrs.	4 hrs.	6 hrs.	9 hrs.	12 hrs.	24 hrs.
		Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.	Per cent.
60	18	22	28	44	66	100	100	100	100	100	100
45	30	10	13	20	33	—	66	87	90	100	100
30	19	5	5	10	44	—	79	94	100	100	100
15	55	5	11	14	33	38	44	71	87	98	100
5	39	0	5	8	15	17	36	54	—	73	98

It is possible that these figures may be somewhat exaggerated, particularly those of the later observations, by reason of the high natural mortality that occurred whilst the experiments were being carried out. On the other hand, the control mortalities generally remained reasonably low until the 12-hour observation.

These results do not altogether agree with those of Knipling (1945), who states that " several hours of continuous exposure (to DDT) are required to cause 100 per cent. knock-down." The results of his tests with *A. quadrimaculatus*, Say, exposed to a 200 mgm. impregnation on wood 4 months after treatment, show that 24 hours after a 60-minute exposure a mortality of only 77 per cent. was obtained, as compared to a 100 per cent. mortality within 3 hours in the tests with *A. gambiae* and *A. funestus* recorded above. This suggests that, either the two African vector Anophelines are more susceptible than *A. quadrimaculatus* to DDT toxicity, or possible differences in the solutions used, or in the mode of application, may have been responsible for the differences of effect.

The results tabulated above indicate that a considerable proportion of *A. gambiae* and *A. funestus* ♀♀ would probably acquire a lethal dose of DDT, after resting for the period normally required for complete oviposition, on a water surface treated at the rate of 200 mgms. DDT per sq. foot.

Comparison of Anopheles resting, in treated and untreated mosquito-net Cages.

Further experiments were carried out in order to determine whether or not DDT impregnation produced any repellent effect on *A. gambiae* or *A. funestus*. For this purpose 1 ft. cube net cages were used, arranged in series of three, of which one cage (A) was impregnated at the rate of 200 mgms. DDT per sq. foot, whilst two untreated cages (B & C) were joined and placed with the entrance opposite to that of the treated cage. Three untreated cages (D, E & F), arranged in a similar manner, were used as a control. The arrangement of the cages is shown diagrammatically.

For the first tests mosquitos were inserted into cages A and D, and after an interval of 5 minutes the entrances x and y were opened to allow free access to cages B-C and E-F respectively. Thereafter observations were made at regular intervals, timed from the opening of x and y. In a second series of tests the mosquitos were inserted into B-C and E-F, and were thus not forced to come into contact with the DDT surface.

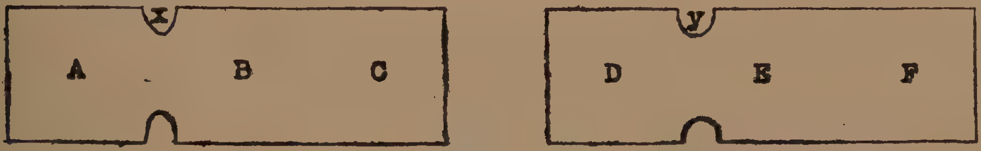

Fig. 1.

Table VII summarises the results of four tests by the first procedure, 50 mixed vector ♀♀ being used in each case.

TABLE VII.

Behaviour of mosquitos having access to both treated and untreated surfaces. (*a*) After 5 minutes enforced contact with a DDT surface.

Time	Per cent. of Total in A.	Per cent. of Total dead in A.	Total mort. Per cent.	Per cent. Flying	Per cent. of Total in D.	Per cent. of Total dead in D.	Total mort. Per cent.	Per cent. Flying
½ hour ...	21	4·5	7	90	43	0·25	0·5	2
1 hour ...	19	11·5	22·5	66	47	0·75	1·0	2
1½ hours ...	12	19	38·5	46	47	1·25	1·5	2
2 hours ...	25	21	51	22	48	1·25	1·5	0
3 hours ...	29	26·5	67	—	44	1·25	2·75	—
Mean in A throughout test, 21 per cent.					Mean in D throughout test, 46 per cent.			

In each of the four tests it was found that there was a considerable restlessness amongst the mosquitos of the A-B-C series, whilst amongst those of the D-E-F series practically no movement took place after the initial disturbance caused by opening the connections x and y had subsided. The numbers in A recorded at each period of observation are thus only approximate. The figures showing the extent of restlessness were only recorded during the fourth experiment, and represent the percentage flying of those that remained alive.

TABLE VIII.

Behaviour of mosquitos having access to both treated and untreated surfaces. (*b*) After no enforced contact with a DDT surface.

Time	Per cent. of Total in A.	Per cent. of Total dead in A.	Total mort. Per cent.	Per cent. Flying	Per cent. of Total in D.	Per cent. of Total dead in D.	Total mort. Per cent.	Per cent. Flying
½ hour	19	0	0	8·5	25	0	0	0
1 hour	11	0	1	14	25	0	0	0
1½ hours	11	0	1	11	24	0	0	0
2 hours	8	1	2	15	25	0	0	0
3 hours	11	3	6	11	26	0	0	0
Mean in A throughout test, 12 per cent.					Mean in D throughout test, 25 per cent.			

Table VIII summarises five trials by the second procedure, in which the mosquitos were not forced to come into contact with the DDT surface. In each trial 40 *A. gambiae* and *A. funestus* ♀♀ were used. Considerable restlessness was again observed in the A-B-C series, and is recorded at each period of observation as a percentage of the total alive.

It would appear from the above tables that the repellent action of a DDT impregnation is negligible, since the mosquitos continued to enter the treated cage, although offered no inducement to do so. The consistently lower figures in the DDT cage in each series of tests may be significant, but it is probable that the degree of difference between the treated and untreated cages is largely due to the restlessness induced by contact with the DDT surface, rather than to any repellent action. The greatly increased restlessness throughout the first tests, in which all the mosquitos used in the treated series had had some initial contact with the DDT surface, indicates a direct connection between contact and restlessness. These results agree with field experiments carried out by Metcalf and others.

General Summary.

1. Certain experiments intended to throw light on problems arising from the practical application of DDT against *A. gambiae* and *A. funestus* are recorded. These refer particularly to the action of larvicidal DDT on adult mosquitos ; to the effect of varying periods of exposure ; and to the possibility of repellence of adults by DDT treatments.

2. (*a*) Solutions of DDT on water surfaces produce an appreciable mortality, particularly on laying mosquitos, but also on ♀♀ in earlier stages of ovary development, when confined in cages with a bowl of water so treated.

(*b*) The greatest effect is produced by a DDT solution in a heavy oil film of long persistency.

(*c*) The presence of a DDT solution on the water, particularly when applied in a heavy oil, appears to have produced an appreciable inhibition against laying.

3. With a 5-months-old residual film of 200 mgms. DDT per sq. foot on smooth wood, a 60-minute exposure killed 22 per cent. immediately and 100 per cent. within 3 hours. After a 5-minute exposure none was killed immediately, but 17 per cent. were dead within 3 hours, and 73 per cent. at 12 hours. Intermediate periods of exposure gave corresponding results.

4. *Anopheles* exposed in inter-connecting treated and untreated cages showed characteristic restlessness as a result of contact with a treated surface. When inserted first into a treated cage, and subsequently allowed access to untreated cages, a higher mortality was recorded than when the procedure was reversed. The degrees of mortality and of restlessness appeared to be closely connected.

References.

Knipling, E. F. (1945). J. Nat. Malar. Soc., **4,** pp. 77–92.

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ASSESSMENT OF MALARIA CONTROL BY MOSQUITO PREVALENCE.*

By K. S. Hocking, A.R.C.S., B.Sc.

Introduction.

The ultimate assessment of the efficiency of malaria control is by the number of cases of malaria contracted in the controlled area, but a method that gives a more immediate and definite information is required because of the :—

(i) Delay before the necessary information is obtained. For example, abandonment of larval control will not affect the number of malaria cases for more than a month ;

(ii) Shifting populations. People will come into the area and develop malaria acquired outside and conversely people having acquired malaria inside the area will go outside before developing it ;

(iii) Difficulty of distinguishing between relapses and newly acquired infections. The number of relapses has no connection with the efficiency of the control ;

(iv) Lack of cases of malaria despite infection in a predominantly immune community in which the non-immune persons are protected by suppressive drugs, mepacrine and quinine. This is a common condition in very malarious areas.

One method which gives more immediate results is the assessment of the efficiency of the control by hand catching of mosquito adults in " Adult Catching Stations " distributed throughout the controlled area and outside. This method has been very widely used by the army in East Africa during the last six years, and it is thought that the conclusions derived from such extensive use are worth recording.

Method.

(*a*) *Type of Station.*—These adult catching stations may be native huts, servants' quarters in a European area, or tents in Military camps, and are usually in groups of 3 or 5.

Where *Anopheles gambiae*, Giles, or *A. funestus*, Giles, are the main vectors concerned, the catching stations must be slept in by human beings as these species are strongly homophilic.

(*b*) *Method of Catching.*—Mosquitos are caught by trained catchers using 6 in. × ½ in. test tubes and an electric torch for illumination. The intention is to catch all the mosquitos in the catching station and this usually takes about 45 mins. per native house. It is probable that generally not more than about half the mosquito population of a native hut was caught (see below).

(*c*) *Time of Catching.*—In 1940 and 1941 these adult catches were made at dawn, but it was shown by Haddow (Bull. ent. Res., **33,** 1942, p. 115) that there was no significant difference in the numbers caught at any time from 6 a.m. until at any rate 11 a.m., and since then catches have been carried out at any time that was convenient during the morning.

(*d*) *Frequency of Catching.*—All adult catching stations were searched once each week. Sometimes all, in one particular control, on one day ; sometimes in six groups, one on each day from Monday to Saturday.

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(*e*) *Distribution of Stations.*—The catching stations were distributed in three belts as follows :—

(i) *Central Belt.*—From the centre of the controlled area to within, say, half a mile of the edge. These catching stations should be completely controlled and assess the efficiency of the control in the area to be protected (*e.g.* town or camp).

(ii) *Marginal Belt.*—These catching stations are outside the town or camp and less than half a mile inside the boundary of the controlled area. They assess mosquito infiltration from outside the area, both in direction and quantity.

(iii) *Extra-Limital Belt.*—These catching stations are outside the controlled area and estimate the mosquito population there, giving by comparison with the central catching stations an assessment of the value of the control measures.

Results illustrating the Value of Hand Catching as a Method of assessing Malaria Control.

TABLE I.

Date	Nairobi			Dar-es-Salaam			Diego Suarez		
	Adult vectors per house	Malaria cases per 1,000	Rainfall in inches	Adult vectors per house	Malaria cases per 1,000	Rainfall in inches	Adult vectors per house	Malaria cases per 1,000	Rainfall in inches
1943									
January	0·02	0·21	0·05			3·2			
February	0	0·7	2·2			9·5	2·0	49	
March ...	0·006	0·83	1·2			2·2	1·6	36	
April ...	0·125	0·74	6·8	2·8	130	14·8	0·6	44	0·5
May ...	0·28	1·8	1·1	3·7	230	10·0	0·5	29	0·2
June ...	0·3	3·85	2·7	4·0	290	0·6	0·2	18	0·4
July ...	0·25	3·21	0·02	2·6	170	2·9	0·1	19	0·9
August...	0·072	1·74	0·4	1·0	40	0·9	0·02	12	0·1
September	0·007	0·56	0·5	1·5	6	1·3	0	12	0·02
October	0·04	0·18	0·4	0·8	9	0·5	0·15	6	0·3
November	0·005	0·2	1·5	1·1	16	0·9		7	0·3
December	0·007	0·35	2·2	0·5	18	1·4	0·12	9	2·3
1944									
January	0·05	0·29	0·3	0·3	19	3·0	0·27	9·3	9·6
February	0	0·37	0	0·3	41	2·2	0·5	10·2	4·3
March ...	0	0·21	5·5	0·6	25	4·3	0·15	7·5	1·1
April ...	0·12	0·37	6·7	6·3	73	14·3	0·23	8·0	2·0
May ...	0·44	0·95	0·9	6·3	150	5·2	0	9·0	0·1
June ...	0·39	1·77	0·9	4·6	174	2·6	0·06	6·0	0·2
July ...	0·07	1·55	0·8	3·2	45	1·3	0	3·8	0·4
August...	0	0·71	0·06	1·8	8	1·2	0·2	2·4	0·2
September	0·01	0·34	2·8	1·2	16	2·1	0	1·0	0·1
October	0·005	0·1	1·4	0·8	5	4·0	0	2·1	0
November	0·045	0·15	5·6	0·8	6	5·4	0	0	0·2
December	0·14	0·26	2·1	2·3	32	2·3	0·2	4·7	6·3
1945									
January	0·12	0·28	1·4	2·0	50	4·8	0·16	1·8	9·3
February	0·03	0·59	2·4	0·7	30	2·2	0·1	4·6	6·6
March ...	0	0·43	2·72	0·4	60	6·0	0·13	0	2·5
April ...	0	0·32	1·24	3·2	70	7·3	0·06	4·5	1·9
May ...	0·005	0·30	9·9	2·3	40	9·2	0·06	0	1·7
June ...	0·05	0·31	2·53	1·8	70	0·4	0·05	0	0·3
July ...	0·05	0·29	1·0	0·3	20	0·4	0·06	3·0	0
August...	0·08	0·24	1·25	0·4	30	0·8	0·08	3·0	1·7
September	0·005	0·3	0·64	0·3	10	1·8			
October	0	0·29	0	0·4	25	4·0			
November	0·01	0·18	6·3	0·5	33	2·4			
December	0·045	0·07	0·6	0·3	8	1·4			

These three widely separated places were selected to illustrate the value of the method because control was carried on in each case for a considerable time and the numbers of malaria cases are reasonably reliable.

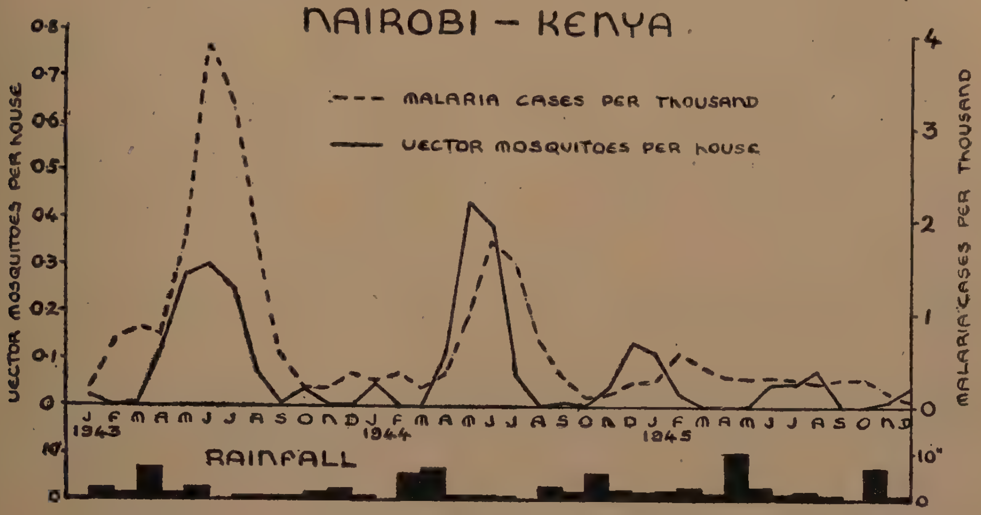

Fig. 1.

In Nairobi the whole of the municipal area was controlled and the population is so large (about 100,000) that small inaccuracies in the number of cases do not affect the general picture.

In Dar-es-Salaam, the aerodrome area was controlled and the malaria cases are those from the R.A.F. station only. These give a true picture of the rate of infection except that after July 1944 personnel were taking suppressive mepacrine and the number of cases was presumably greatly reduced.

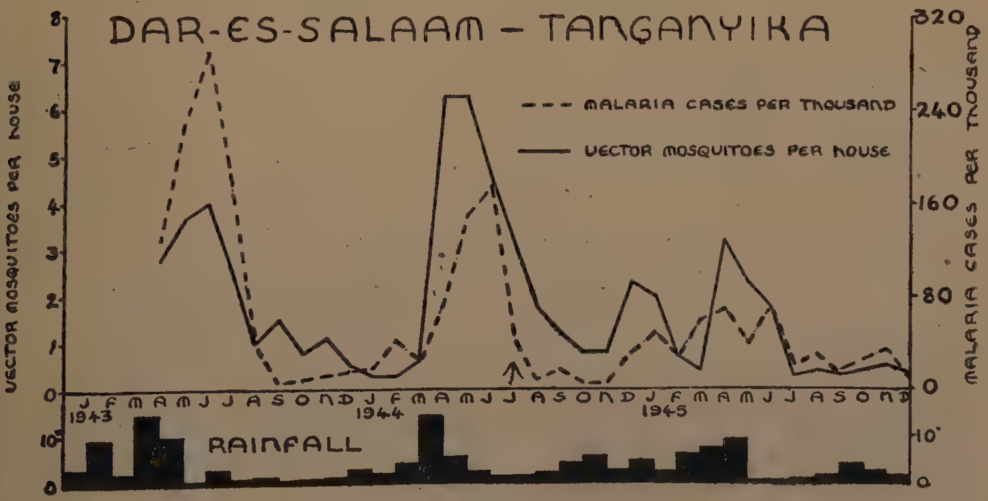

Fig. 2.

The Diego Suarez figures are for Europeans of the Army and R.A.F. and probably give a fairly true picture of the rates of infection from the start of control through two rainy seasons.

The graphs (figs. 1–3) derived from these figures show that generally the number of cases of malaria follows the number of vector mosquitos per house ; each appreciable rise in the number of cases of malaria is foretold by a rise in the adult catches.

In Dar-es-Salaam and Diego Suarez the correlation is greatest between adult catches and malaria cases two weeks later (actually the mean of the cases for the current month and the following month), and in the cooler climate of Nairobi between adult catches and malaria cases one month later. The respective maximum correlation coefficients for the three places are 0·69, 0·85 and 0·77 ; all highly significant.

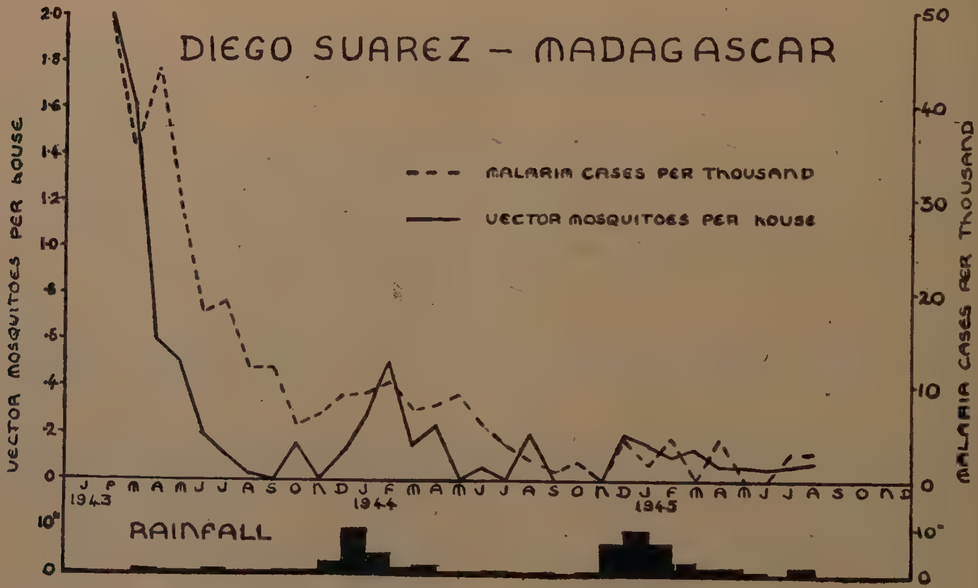

Fig. 3.

Hence from the point of view of assessing the efficiency of the control, the adult catches give information as accurate as, and more immediate than, the number of malaria cases.

TABLE II.

Weekly Mosquito Catches from Dar-es-Salaam to illustrate central, marginal and extra-limital Catches.

Week ending	Average of 12 Central Stations	Average of 15 Marginal Stations	Average of 9 Extra-Limital Stations
14th April, 1945 ...	0·2	7·0	15·4
21st " " ...	0·8	9·7	20·1
28th " " ...	2·7	8·4	16·0
5th May, 1945 ...	0·9	6·1	14·1
12th " " ...	0·3	8·0	16·1
19th " " ...	0·9	9·6	23·2
Mean	0·97	8·1	17·5

Comparison of the mean central and extra-limital catches given above suggests that the control of the *Anopheles* in the central area is about 95 per cent. efficient. The marginal catches, as was to be expected from stations protected on one side only, averaged about half the extra-limital catches. The only marginal station with catches much higher than this was M1, in the N.W. corner of the controlled area (average 25·1), indicating that the greatest infiltration of mosquitos at this time was from the N.W. direction.

Disadvantages of Method.

(*a*) It is not always possible to find suitable catching stations in the places required.

(*b*) Searching in native huts is difficult and, therefore, the number of mosquitos caught depends on the efficiency of the searcher.

(*c*) Native huts vary very much from place to place, both in size and suitability for mosquito harbourage, so catches per hut are not comparable from place to place.

(*d*) Tents used as catching stations become unsuitable for mosquitos in the heat of the day and an unknown proportion leave during the morning.

(*e*) A very small Anopheline population may not be represented in weekly searches in any reasonable number of catching stations.

One plausible attempt to avoid disadvantage (*b*) is by replacing hand catching of the mosquitos by spraying the huts with pyrethrum extract and collecting the dead mosquitos from sheets previously spread on the floor. This method undoubtedly produces larger numbers—in our experience generally about twice as many (probably chiefly because of mosquitos resting in positions such as the roof, inaccessible to hand catchers)—but most native huts are far from mosquito proof and a large and unknown number escapes during spraying. Also it is rarely possible to spread sheets satisfactorily in a furnished native hut and consequently the number collected probably bears no more definite relation to the total number present.

A Possible Improvement.

In an attempt to overcome these disadvantages, an experimental type of standardised portable trap hut was introduced in 1945. This was of wooden frame construction 6 ft. high and 8 ft. by 4 ft. 6 ins. inside. Around it, 3 ft. 6 ins. up, were panels of gauze 1 ft. deep, above and below which were vertical slits protected by an overhanging flap of gauze.

The theoretical advantages of portable standardised trap huts are :—

(i) They can be erected singly or in groups wherever required.

(ii) Mosquitos are comparatively easy to find in them.

(iii) They are standardised and catches in them are comparable from place to place.

(iv) Mosquitos entering them are trapped and therefore the weekly catch is an accumulation from seven nights' entry.

In practice, unless great care was taken with the experimental model to ensure the proper adjustment of the gauze flanges, the closing of the door, and the mending of all cracks appearing in the walls, mosquitos were not trapped.

During the latter part of 1945 these traps were tried out in several areas, particularly in the Port Reitz area near Mombasa, and the military camp area near Moshi, Tanganyika. After numerous preliminary difficulties, particularly in persuading people to sleep in them, the traps started giving what appeared to be valuable results.

Table III compares the catches per trap and the catches per native hut in the various zones of the controls at Mombasa and Moshi for August to December, 1945.

TABLE III.

Place			Aug.	Sept.	Oct.	Nov.	Dec.	Mean
Mombasa ...	Central Zone ...	Trap ...	0·1	0·02	0	0	0	0·02
		Hut ...	0	0	0	0	0·015	0·003
	Marginal Zone ...	Trap ...	0·8	0·4	0·05	0	0	0·25
		Hut ...	0·3	0·05	0·02	0	0	0·07
	Extra-Limital Zone	Trap ...	6·7	1·2	0·8	0·05	0·24	1·8
		Hut ...	12·6	10·8	7·8	5·5	3·9	8·1
Moshi ...	Central Zone ...	Trap ...		0·1	0	0·04	0	0·04
		Hut ...		0	0	0·03	0·13	0·04
	Extra-Limital Zone	Trap ...		0·3	0·2	0·3	1·4	0·55
		Hut ...		0·1	1·5	0·8	0·55	0·74

Because some traps were not slept in regularly, and some only twice a week by askaris under nets, the catches from them are not as high as they might be; but since, when mosquitos are numerous, as in the extra-limital areas, the trap catches are lower than the native hut catches, it would seem that mosquitos are escaping from the traps.

Investigations are in progress to find ways of improving these trap-huts.

Conclusions.

1. Routine hand catching of adult mosquitos gives a prompt indication of any deficiency in a control.

2. The adult catches generally give as reliable an assessment of the efficiency of a control as do the cases of malaria.

3. Probably greater precision can be obtained by the use of portable trap huts as standardised catching stations.

INSECT PESTS OF SOME ECONOMIC CROPS IN FIJI. No. 2.

By R. J. A. W. Lever, B.Sc., D.I.C., A.I.C.T.A., F.L.S.

1. CROPS.

Ananas comosus (Pineapple).
Pycnoscelus surinamensis, L. (Blattid.)—Leaf.

Annona reticulata (Custard Apple).
Aspidiotus destructor, Sign. (Coccid.)—Leaf.
Ferrisia virgata, Ckll. (Coccid.)—Fruit.
Pseudococcus citri, Risso (Coccid.)—Fruit.

Artocarpus incisa (communis) (Breadfruit).
Dacus passiflorae, Frogg. (Trypetid.)—Fruit.

Avocado see *Persea*.

Banana see *Musa*.

Bean see *Phaseolus*.

Bele see *Hibiscus*.

Beta vulgaris (Beet).
Nezara viridula, L. var. *smaragdula*, F. (Pentatomid.)—Young leaf.

Brachiaria mutica (Para Grass).
Marasmia venilialis, Wlk. (Pyralid.)—Leaf (roller).
Mocis trifasciata, Steph. (Noctuid.)—Leaf.
Remigia frugalis, F. (Noctuid.)—Leaf.

Brassica campestris (Turnip).
Brassica chinensis (Chinese Cabbage).
Liriomyza strigosa, Mg. (Agromyzid.)—Leaf (miner).

Breadfruit see *Artocarpus*.

Cabbage see *Brassica*.

Cajanus cajan (Pigeon Pea, Dahl).
Maruca testulalis, Geyer (Pyralid.)—Pod.

Camellia sinensis (Tea).
Icerya seychellarum, Westw. (Coccid.)—Leaf, Twig.

Capsicum fragrans (Chilly).
Dacus (Chaetodacus) passiflorae, Frogg. (Trypetid.)—Fruit.
Icerya seychellarum, Westw. (Coccid.)—Twig.
Nezara viridula var. *smaragdula*, F. (Pentatomid.)—Fruit, Stem.

Carica papaya (Pawpaw).
Elytrurus griseus, Guér. (Curculionid.)—Leaf.

Cassia occidentalis.

Leptoglossus australis, F. (Coreid.)—Stalk, Pod.
Mictis profana, F. (Coreid.)—Pod.
Nezara viridula var. *smaragdula*, F. (Pentatomid.)—Pod.
Protaetia fusca, Hbst. (Cetoniid.)—Leaf, Pod.

Castor see *Ricinus*.

Chestnut, Tahitian see *Inocarpus*.

Chilly see *Capsicum*.

Citrus limonia (Lemon).
Citrus nobilis (Mandarin).
Citrus sinensis (Orange).
Citrus aurantiifolia (Lime).

Aonidiella aurantii, Mask. (Coccid.)—Stem, Leaf, Fruit.
Ceroplastes rubens, Mask. (Coccid.)—Twig.
Fiorinia proboscidaria, Green (Coccid.)—Leaf.
Icerya seychellarum, Westw. (Coccid.)—Leaf.
Dacus xanthodes, Broun (Trypetid.)—Fruit.
Papilio schmeltzi, H.-S. (Papilionid.)—Leaf.
Elytrurus griseus, Guér. (Curculionid.)—Leaf.

Cocoa see *Theobroma*.

Cocos nucifera (Coconut).

Batrachedra arenosella, Wlk. (Elachistid.)—Male flowers.
Pyroderces paroditis, Meyr. (Elachistid.)—Male flowers.
Rhabdoscelus (*Rhabdocnemis*) *obscura*, Boisd. (Curculionid.)—Trunk.
Rhopaea vestita, Arrow (Rutelid.)—Leaf.
Pinnaspis minor, Mask. (Coccid.)—Leaf.

Colocasia esculenta (Dalo, Taro).

Agrotis ypsilon, Hb. (Noctuid.)—Leaf.
Brachylybas variegatus, Le Guill. (Coreid.)—Stem.

Cotton see *Gossypium*.

Cowpea see *Vigna*.

Cucurbita pepo (Marrow, Pumpkin).

Aulacophora quadrimaculata, F. (Galerucid.)—Leaf.
Brachylybas variegatus, Le Guill. (Coreid.)—Stem.
Plusia chalcites, Esp. (Noctuid.)—Leaf.
Prodenia litura, F. (Noctuid.)—Leaf.

Custard Apple see *Annona*.

Dahl see *Cajanus*.

Dalo see *Colocasia*.

Egg-plant see *Solanum*.

Ginger see *Zingiber*.

Gossypium barbadense (Sea Island Cotton).
Ferrisia virgata, Ckll. (Coccid.)—Stem.
Pinnaspis minor, Mask. (Coccid.)—Stem.
Saissetia nigra, Nietn. (Coccid.)—Stem.
Anomis vulpina, Butler (Noctuid.)—Leaf.
Plusia chalcites, Esp. (Noctuid.)—Leaf.
Platyedra gossypiella, Saund. (Gelechiid.)—Boll.
Rhinoscapha lagopyga, Frm. (Curculionid.)—Leaf.

Gourd, Bitter see *Momordica*.

Gourd, Bottle see *Lagenaria*.

Gourd, Snake see *Trichosanthes*.

Granadilla see *Passiflora*.

Hibiscus manihot (Bele).
Anomis flava, F. (Noctuid.)—Leaf.
Earias fabia, Stoll (Noctuid.)—Bud, Boll.
Sylepta derogata, F. (Noctuid.)—Leaf.
Elytrurus griseus, Guér. (Curculionid.)—Leaf.

Inocarpus edulis (Ivi, Tahitian Chestnut).
Strumeta passiflorae, Frogg. (Trypetid.)—Fruit.
Prodenia litura, F. (Noctuid.)—Fruit.

Ipomoea batatas (Sweet Potato).
Nezara viridula var *smaragdula*, F. (Pentatomid.)—Vine.
Liriomyza strigosa, Mg. (Agromyzid.)—Leaf.
Ercta ornatalis, Dup. (Pyralid.)—Leaf.

Ivi see *Inocarpus*.

Lactuca sativa (Lettuce).
Thrips tabaci, Lind. (Thysanopt.)—Leaf (crinkle).
Nezara viridula var. *smaragdula*, F. (Pentatomid.)—Leaf.
Heliothis armigera, Hb. (Noctuid.)—Leaf.
Prodenia litura, F. (Noctuid.)—Leaf.

Lemon see *Citrus*.

Lettuce see *Lactuca*.

Lime see *Citrus*.

Lagenaria leucantha (Bottle Gourd).
Luffa acutangula.
Luffa cylindrica (Luffa).
Leptoglossus australis, F. (Coreid.)—Fruit.

Lycopersicum esculentum (Tomato).
Brachylybas variegatus, Le Guill. (Coreid.)—Stem.
Dacus xanthodes, Broun (Trypetid.)—Fruit.

Maize see *Zea*.

Mandarin see *Citrus*.

Mangrove see *Rhizophora*.

Marrow see *Cucurbita*.

Mimosa pudica (Sensitive Plant).
Icerya seychellarum, Westw. (Coccid.)—Stem.

Miscanthus japonicus.
Graeffea crouani, Le Guill. (Phasmid.)—Leaf.

Momordica charantia (Bitter Gourd).
Leptoglossus australis, F. (Coreid.)—Fruit.

Musa cavendishii (Chinese Banana).
Musa paradisiaca (Plantain).
Musa sapientum (Banana).
Stenomatus musae, Mshl. (Curculionid.)—Stem.
Prodenia litura, F. (Noctuid.)—Leaf.
Aspidiotus destructor, Sign. (Coccid.)—Leaf.

Nicotiana tabacum (Tobacco).
Nezara viridula var. *smaragdula*, F. (Pentatomid.)—Stem.

Orange see *Citrus*.

Oryza sativa (Rice).
Marasmia venilialis, Wlk. (Pyralid.)—Leaf (roller).

Pandanus tectorius.
Cardiodactylus novae-guineae, De Haan (Gryllid.)—Leaf.
Nacoleia octasema, Meyr. (Pyralid.)—Flowers.

Para Grass see *Brachiaria*.

Passiflora quadrangularis (Granadilla).
Prodenia litura, F. (Noctuid.)—Leaf.
Alciphron glaucus, F. (Pentatomid.)—Fruit.
Brachylybas variegatus, Le Guill. (Coreid.)—Stem.

Pawpaw see *Carica*.

Persea americana (Avocado Pear).
Dacus (*Chaetodacus*) *passiflorae*, Frogg. (Trypetid.)—Fruit.
Aspidiotus destructor, Sign. (Coccid.)—Leaves, Twigs.

Phaseolus calcaratus (Rice Bean).
Aphis gossypii, Glov. (Aphid.)—Shoots, Buds.

Phaseolus lunatus (Lima Bean, Madagascar Bean).
Phaseolus vulgaris (French Bean).
Elytrurus griseus, Guér. (Curculionid.)—Leaf.
Plusia chalcites, Esp. (Noctuid.)—Leaf.
Melanagromyza phaseoli, Coq. (Agromyzid.)—Leaf (miner).

Pigeon Pea see *Cajanus*.

Pineapple see *Ananas*.

Piper methysticum (Yagona).
Saissetia nigra, Nietn. (Coccid.)—Stem.
Elytrurus griseus, Guér. (Curculionid.)—Leaf.

Plantain see *Musa*.

Potato see *Solanum*.

Pumpkin see *Cucurbita*.

Raphanus sativus (Radish).
Myzus persicae, Sulz. (Aphid.)—Leaf.
Rhopalosiphum pseudobrassicae, Davis (Aphid.)—Leaf.

Rhizophora mangle (Mangrove).
Aspidiotus suvaensis, Green (Coccid.)—Stem, Leaves.

Rice see *Oryza*.

Ricinus communis (Castor).
Thalassodes pilaria, Guen. (Geometrid.)—Leaf.
Nezara viridula var. *smaragdula*, F. (Pentatomid.)—Flowers, Leaves.

Sacchárum officinarum (Sugar-cane).
Perkinsiella saccharicida, Kirk. (Delphacid.)
Decadarchis flavistriata, Wlsm. (Lyonetiid.)—Young buds.
Adoretus versutus, Har. (Rutelid.)—Roots.
Lacon stricticollis, Frm. (Elaterid.)—Young shoots, Roots.
Simodactylus cinnamomeus, Boisd. (Elaterid.)—Stools.

Sensitive Plant see *Mimosa*.

Solanum melongena (Egg-plant).
Spodoptera mauritia, Boisd. (Noctuid.)—Leaf, Fruit.
Adoretus versutus, Har. (Rutelid.)—Leaf.

Solanum torvum.
Prodenia litura, F. (Noctuid.)—Leaf.
Epilachna vigintioctopunctata, F. (Coccinellid.)—Leaf.

Solanum tuberosum (Potato).
Agrotis ypsilon, Hfn. (Noctuid.)—Leaf.

Sugar-cane see *Saccharum*.

Sweet Potato see *Ipomoea*.

Tamarindus indica (Tamarind).
Pachymerus gonager, F. (Bruchid.)—Seeds.

Taro see *Colocasia*.

Tea see *Camellia*.

Theobroma cacao (Cocoa).
Pseudococcus citri, Risso (Coccid.)—Pod.
Dacus passiflorae, Frogg. (Trypetid.)—Fruit.
Prodenia litura, F. (Noctuid.)—Leaf.

Tobacco see *Nicotiana*.

Tomato see *Lycopersicum*.

Trichosanthes anguina (Snake Gourd).

Leptoglossus australis, F. (Coreid.)—Fruit.

Turnip see *Brassica*.

Urena lobata.

Pseudaulacaspis pentagona, Targ. (Coccid.)—Stalk.
Anomis vulpina, Butl. (Pyralid.)—Leaf.

Vigna sinensis (Cowpea).

Aphis gossypii, Glov. (Aphid.)—Leaves, Stem.
Leptoglossus australis, F. (Coreid.)—Leaf, Stalk, Pod.
Lygus muiri, Popp. (Mirid.)—Leaf.
Adoretus versutus, Har. (Rutelid.)—Leaf.

Yagona see *Piper*.

Zea mays (Maize):

Leptoglossus australis, F. (Coreid.)—Young seeds.
Nezara viridula var. *smaragdula*, F. (Pentatomid.)—Cob.
Marasmia trapezalis, Guen. (Pyralid.)—Leaf (roller).
Nacoleia octasema, Meyr. (Pyralid.)—Leaf.
Rhopaea vestita, Arrow (Rutelid.)—Roots.

Zingiber officinalis (Ginger).

Adoretus versutus, Har. (Rutelid.)—Leaf.

2. STORED PRODUCTS.

Leguminous Seeds.

Bruchus maculatus, F. (Bruchid.).
Bruchus pisorum, L. (Bruchid.).
Bruchus tristiculus, Fhs. (Bruchid.).
Pyralis pictalis, Dup. (Pyralid.).

Maize Meal and Derris.

Tenebroides mauritanicus, L. (Trogositid.).

Rubbish Feeder.

Ahasverus advena, Waltl (Cucujid.).

3. MISCELLANEOUS MATERIALS.

Bamboo Counters & Leather.

Catorama herbarium, Gorh. (Anobiid.).

Bat Guano.

Carcinops 14-*striata*, Steph. (Histerid.).
Euxestus parki, Woll. (Colydiid.).
Monopis strichomela, Low. (Tineid.).
Atherigona excisa, Thoms. (Anthomyiid.).
Atherigona excisa trilineata, Stein (Anthomyiid.).
Desmometopa ciliata, Hend. (Milichiid.).
Milichia angustifrons, Bezzi (Milichiid.).

Carcases (Sheep, Cattle).

Necrobia ruficollis, F. (Clerid.).
Necrobia rufipes, Deg. (Clerid.).

Decaying Plant Tissue.

Atherigona excisa, Thoms. (Anthomyiid.).
Megaselia scalaris, Lw. (Phorid.).

Felt.

Anthrenus pimpinellae, F. (Dermestid.).

Scavenger.

Anoplolepis longipes, Jerd. (Formicid.).

Tobacco-Pyrethrum Mixture & Ice Cream Cones.

Lasioderma serricorne, F. (Anobiid.).

CORRECTIONS TO LIST I (*Bull. ent. Res.*, **35**, 1945, pp. 367–377).

Transferred to text. E.M.N.

Page 370, line 2, for "*Graeffea cocophaga*, Newp." (on *Cocos nucifera*) read "*Graeffea crouani*, Le Guill."
,, 370, lines 19, 21, for "*Aulacophora coffeae*, Hornstedt" (on *Cucumis sativus* and *Cucurbita pepo*) read "*Aulacophora similis*, Oliv."
,, 370, lines 33, 34, the two Coccids on *Dioscorea* are found on tubers.
,, 371, line 15, for "*Empoasca quadripunctata*, Evans" (on *Gossypium barbadense*) read "*E. quadripunctata*, Lever"
,, 373, line 23, *Nacoleia diemenalis*, Guen. (on *Phaseolus*) is a leaf roller.
,, 374, line 19, delete *Brachyplatys pacificus*, Dall. (on *Saccharum officinarum*).

THE BIOLOGY OF THE PARASITES OF *LOXOSTEGE STICTICALIS*, L., IN NORTH AMERICA—*BRACON VULGARIS* (CRESS.) (BRACONIDAE, AGATHINAE).

By F. J. Simmonds.
Imperial Parasite Service. E.H.N.

Introduction.

In a previous paper (Simmonds 1946*) the biology of *Phytodietus pulcherrimus* (Cress.), a parasite of *Loxostege sticticalis*, L., the sugar-beet web-worm, has been described. This paper will be devoted to the habits and life-history of another important parasite of *Loxostege* in North America, *Bracon vulgaris* (Cress.). This species appears to be confined to the American continent, all of the records being from Canada and the U.S.A. It has been reared only from *Loxostege sticticalis*, L., and *L. commixtalis*, Wlk. From the former species it has been recorded by Paddock (1912), Swenk (1918), Jones, Hoerner and Corkins (1921), Pepper and Hastings (1941), and Simmonds (1946); from the latter species by Hoerner (1933). During the present work it has been reared from *L. sticticalis*, material from both Montana and Alberta.

It is a common parasite of *L. sticticalis*, and its range is probably co-extensive with that of the host, throughout the Great Plains and Rocky Mountains in North America. In the present investigations, hibernating material was parasitised by *Bracon* to the extent of 6·4 per cent. in 1941, 8·0 per cent. in 1942 and 5·5–8·4 per cent. in 1945.

Both Paddock and Swenk state that *Bracon* acts as an effective check on the increase of *L. sticticalis*, and both refer to the decrease in its effectiveness caused by the secondary parasite, *Mesochorus agilis*, Cress. *Mesochorus perniciosus*, Vier., was reared as a secondary parasite of *Loxostege* during the present work (possibly in actual fact the same species referred to by Paddock and Swenk), its immediate host being *Bracon* in the majority of instances. From the emergence records it was seen to destroy 41·7 per cent. of the primary parasites in 1941, 18·8 per cent. in 1942, and about 5·6 per cent. in 1945. The secondary parasite, *Perilampus similis*, Cwfd., was also reared in small numbers, and sometimes *Bracon* was its immediate host.

Parasitism of *Loxostege* by *Bracon* has been recorded as high as 98 per cent. in a collection of larvae made in Montana in 1935 (Pepper and Hastings 1941). In two other collections of the same year 17 per cent. and 26 per cent. of the host larvae were parasitised. They also state "in 1936 the first generation larvae collected in and around Bozeman, Montana, were 26 per cent., and the second generation larvae at Great Falls were 8 per cent. parasitised. Although no further counts were made in later years, heavy parasitism has been noted occasionally in localized areas." The most common species of parasite in these collections was *Bracon vulgaris* and, in sweepings made over infested areas, the ratio of *Bracon* to all the other parasites was 50 to 1. It is thus apparent that *Bracon* is an important parasite of *Loxostege* in all the collections made.

Systematic Position and Adult Characters.

Bracon vulgaris (Cress.) belongs to the subfamily AGATHINAE of the family Braconidae. According to Morrison (1917) the older classifications (e.g. that of Marshall) included the sub-family with the MICROGASTERINAE in the group Areolarini of Wesmael (=Areolarii of Schmiedekneckt), but the classifications of Ashmead (1900) and Szépligeti (1904) present the sub-families without grouping them into higher units.

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*In course of publication.

The species was originally described by Cresson in 1865 and placed in the genus *Agathis*, but has since been transferred to the genus *Bracon* Fabricius (=*Cremnops* Foerster *et auct.*). The synonymy as given by Morrison is :—

Agathis vulgaris	Cresson, Proc. ent. Soc. Philad., **4**, 1865, p. 295.
Agathis media	Cresson, Proc. ent. Soc. Philad., **4**, 1865, p. 295.
Agathis exoratus	Cresson, Trans. Amer. ent. Soc., **4**, 1872, p. 182.
Cremnops vulgaris	Ashmead, Bull. Colo. Biol. Ass. no. 1, 1890, p. 41.
Disophrys vulgaris	Marsh, U.S. Dep. Agric., Bur. Ent., Bull. 109, pt. 6, 1912, p. 62.

The adult was originally described by Cresson in 1865, and a very complete description from a study of 350 specimens was given by Morrison (1917) in his monograph on the revision of the genus *Bracon*. In a more brief description he says " color mostly red, orange-yellow, or honey-yellow, head more or less black on the cheeks, venter black, propodaeum black in the male. Length averaging about 8 mm., but varying from 6·5–9·5 mm., Wings—length, average, fore—7·5 mm., hind—5·5 mm., fuliginous." In the female the ovipositor is " pale red, valves blackish, about as long, or when fully retracted, a little shorter than the abdomen." This short description is sufficient to distinguish the species from other parasites reared from *Loxostege* in North America.

General Biology.

In hibernating *Loxostege* material *Bracon* is present as the first stage larva within the host, which is in diapause within its spin-up. When the biochemical changes that precede the beginning of pupation of the host take place the parasite larva feeds and grows rapidly, destroying the host larva ; in due course it forms a cocoon and emerges at about the same time as moths from unparasitised hosts. According to Pepper and Hastings (1941) the males emerge in the field at the same time as the adult *Loxostege* and feed on the blossoms of fan-weed, golden-rod, etc. ; the females are not found until 7–12 days later. Material placed at 85°F. in the laboratory, and " forced " out of diapause, gave an average emergence date for female *Bracon* only one day later than for the males. In 1941, 37·9 per cent. of the emergent *Bracon* from hibernating material were females, in 1942, 29·2 per cent. and in 1945, 28·4 per cent. Of 162 specimens bred in the laboratory 60, or 37·0 per cent., were females, so it would seem that this preponderance of males is of general occurrence.

In the laboratory adults were kept at 75°F. in cheese-cloth sided and celluloid-fronted cages, and supplied with water, raisins and sugar lumps. Length of life was very variable and seemed to depend partly on the treatment of the hibernating material prior to emergence. Laboratory bred material gave an average length of life of approximately 17 days for both sexes.

Mating occurs readily on emergence. Males are very active, and become excited even by the proximity of another male, with which they will often attempt to mate. When a female is found the male pursues her, and endeavours to mount on her back with the tip of the abdomen bent under that of the female, who invariably attempts to shake the male off. When the male is successful in retaining his hold, copulation, which is of very short duration, takes place, and the male is finally shaken off by the female.

When the parasite cocoons have been kept at 85°F. for some time prior to emergence the females may commence ovipositing immediately they leave the cocoon. Mating is not a necessary preliminary to oviposition, unfertilised eggs producing male offspring. Females have been seen to emerge, walk on to an infested beet leaf, and immediately begin stabbing at the host larvae ; and from these larvae adult male *Bracon* have been reared. However, when the cocoons have been kept at 75°F. there is apparently a pre-oviposition period of about a week in which no eggs are laid, and during which little interest is evoked by the presence of host larvae or traces of them. It seems probable that this difference is connected with the rate of maturation of the eggs within the ovaries, which may reach complete maturity before emergence in females from cocoons kept at 85°F. but not in those emerging at the lower temperature.

When the oviposition period has commenced, any trace of silk or frass from the larvae of *Loxostege* excites the female. The spot is tapped violently with the antennae which are curved down parallel to one another almost touching at the tips, which are pressed against the infested surface and vibrated quickly.

This antennal exploration is often followed by stabbing movements with the ovipositor. The abdomen is bent downwards and forwards, the ovipositor is partially freed from the enveloping sheath, and is extended so that its tip comes under the parasite's head and touches the surface near the tips of the antennae. The ovipositor stabs at random over places where traces of host larvae occur, until a host larva is touched by its tip. The stabbing then becomes more excited and the ovipositor is driven into the host, which in some cases is held down by the fore-legs of the parasite and prevented from wriggling away. If the larva succeeds in moving, the ovipositing *Bracon* is dragged along with it, the ovipositor remaining in the larva until the egg is laid. The time taken before the egg is actually laid varies from a few seconds to about a minute, no doubt being dependent, amongst other things, on the number of fully developed eggs contained in the oviducts ready for oviposition. When an egg has been laid the ovipositor is withdrawn and the parasite moves off, obviously no longer interested in the host larva. This behaviour contrasts with that following an unsuccessful attack, when the parasite is still very excited on recontacting the host. Dissections show that only one egg is usually deposited at a time, but it is possible that two may occasionally be laid. However, never more than one adult parasite emerges from a host.

Loxostege larvae that have just emerged from the egg are readily attacked in the laboratory. These are so small that occasionally after oviposition they remain attached to the ovipositor and are pushed off by the *Bracon* with its hind legs. However, these small larvae feed normally after such an attack and produce adult parasites. Fully grown larvae are also attacked, but they are so active and powerful that they can usually escape by moving off more quickly than the *Bracon* can pursue them. In the field when these larvae are in silk webbing it is possible that they will be successfully attacked, and in the laboratory oviposition occurs if the large larvae are held down while the parasite attacks them ; from these larvae progeny can be successfully reared. Females show no interest, however, in hibernating larvae removed from their cocoons and will not parasitise them.

Hence larvae of all sizes can be successfully attacked by *Bracon*, but the most suitable for use in the laboratory for propagation are half-grown larvae about 1 cm. in length. These the *Bracon* attack, and hold down easily with the forelegs while oviposition occurs. Paddock (1912) states that pupae are also attacked, but this has not been found to occur, and certainly it would appear extremely unlikely that this occurs in the field in view of the location of the host pupae within cocoons in the soil.

Laboratory Breeding.

To obtain material for shipment to South Africa a cage was used with three sides and the bottom of wood, a glass top, and the fourth side (6 in. broad × 2 in. high) plugged with cotton wool (the cage being 6 in. × 2 in. × 3 in.). Into this cage four or five ovipositing *Bracon* females were placed, and an electric lamp placed over the cage to keep the parasites active. Suitable host larvae were then introduced singly into the cage over the cotton wool plug and removed with a camel-hair brush after they had been successfully attacked. Whether there is any avoidance of superparasitism or whether a host that has been attacked is just as acceptable to the ovipositing female as a healthy host was not determined.

The number of eggs laid by individual females varies considerably. Sometimes none is laid, though the females are apparently otherwise normal. The maximum recorded was 258 from a particularly large and robust female. The eggs were laid from the day of emergence (the female had emerged at 85°F.) regularly for eight days, after which a reduced number were laid for 4 days and the female then lived for a further six days without laying.

Three other females gave respectively, 122, 25 and 25 eggs. These females emerged at 75°F. and had preoviposition periods in which no eggs were laid. The one that laid 122 eggs had a two-day preoviposition period and laid most of its eggs between the 11th and 15th days. Of the two that laid 25 eggs one laid on one day only—the 14th after emergence—the other laid on the 11th–14th days. It is impossible to give from these figures an average number per female, but they do indicate the variation in numbers of eggs laid.

Description of the Immature Stages.

(*a*) *The Egg.*

At the moment of oviposition the egg is minute (fig. 1, *a*) (about ·16 mm. × ·07 mm.), tear-shaped, and enclosed in a soft membraneous chorion. It is laid free in the body cavity of the host and gradually becomes larger, presumably by osmosis or active absorption of fluid through the egg membranes. After three days at 85°F. the egg is oblong and measures about ·2 mm. × ·13 mm., the "stalk" being reduced to a small projection at one side. After 4 days the egg is oval and ·3 mm. × ·18 mm., still with a slight projection. On the 5th day it measures about ·6 mm. × ·6 mm.,

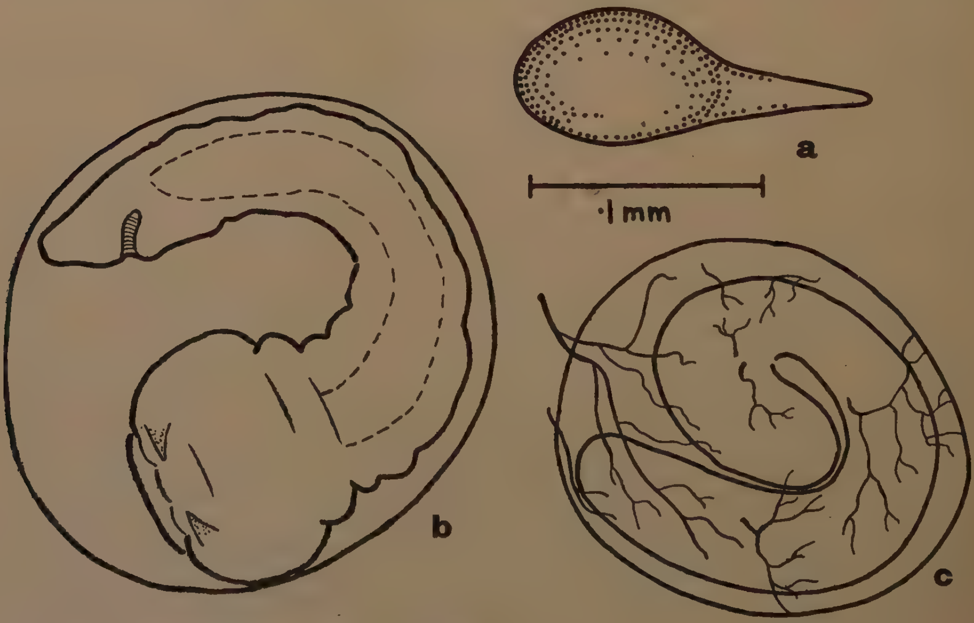

Fig. 1.—*Bracon vulgaris* (Cress.). (*a*) Freshly laid egg ; (*b*) Egg just prior to eclosion ; (*c*) Egg just prior to eclosion, showing host tracheids.

and contains a well developed larva, with head capsule and mandibles distinct, the whole egg being now a soft gelatinous sphere, about 350 times the volume of the original egg. Fig. 1, *b* shows the growth of the egg before eclosion. The volume of the egg at laying, the 3rd, 4th, and 5th days are in the ratio of approximately 1 : 5 : 18 : 350 (see fig. 2).

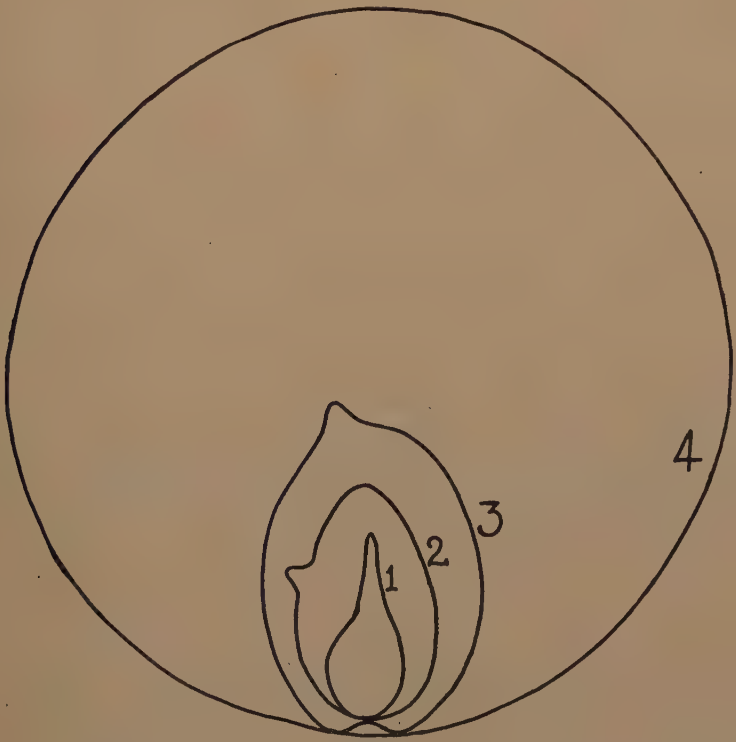

Fig. 2.—*Bracon vulgaris* (Cress.). Comparative growth of the egg before eclosion. 1. Size at oviposition ; 2. 3rd day ; 3. 4th day ; 4. 5th day.

The great increase in the size of the egg between oviposition and eclosion suggests that the membrane surrounding the egg has some trophic properties, absorbing nutriment for the developing embryo from the surrounding body-fluid of the host. Such egg-membranes are found fairly commonly among endoparasitic Hymenoptera, the method of origin varying in different cases. In some instances the membrane is formed by cells arising from the division of the polar nuclei of the egg ; this occurs particularly in polyembryonic forms. In some Chalcids such a membrane is formed by delamination from the blastoderm, and *Litomastix* has one membrane formed by delamination and one from the polar nuclei. In the Braconid, *Diaeretus rapae*, Curt. Spencer (1926) has studied the formation of such embryonic membranes and states that a membrane is formed by delamination and extrusion of cells from the embryonic ectoderm. This membrane he calls a serosa, which is misleading since a true serosa has a different origin in the undifferentiated blastoderm. From the description given by Spencer it would appear that the membrane formed in the case of *Bracon vulgaris* is similar to that of *Diaeretus*, but its origin has not been investigated. Hence it is advisable to term it merely a trophic membrane, since the terms amnion and serosa are restricted to structures with a definite embryological derivation, and

Imms (1937) restricts the term trophamnion to a membrane derived from the polar bodies. In the present work similar increase in the size of the egg has been found to occur with *Meteorus loxostegei*, Vier., and Jackson (1928) records an increase of over 1,200 times in the volume of the egg of *Dinocampus rutilus*, Nees.

On the fifth day the egg is often found adhering to portions of the fat-body, and host tracheids may be found spreading over its surface (fig. 1, *c*) although this does not occur in every case. Presumably the high rate of metabolism occasioned by the developing parasite larva within the egg membranes induces the formation of tracheoles by the tracheal epithelium of the host. A similar activation of the tracheal epithelium due to metabolic activity of a parasite larva has been suggested by Thorpe (1936) in the case of *Encyrtus infelix* (Embleton), a parasite of *Coccus hesperidum*, L.

(*b*) *First-stage Larva.*

On hatching the larva is of a caudate polypodeiform type, curved towards the ventral side, lying in the host body cavity. There are thirteen body segments tapering to the posterior end, where there is a short tail. On each of segments 2–12 there is a pair of bifid ventro-lateral projections, which are less well developed anteriorly and posteriorly than they are on the intervening segments. The significance of both " tail " and ventral appendages is uncertain, and their function is obscure. The skin is smooth and has no definite sculpturing.

The tracheal system is very poorly developed, and although short spiracular branches were seen, they appeared to be " blind ", no spiracles being present.

The head capsule (fig. 3) is about ·25 mm. in width, and is of the ventrally unchitinised Braconid type. The mandibles and general head capsule are well chitinised, and there are raised granulated areas at the ventral edges of the lateral parts of the head shield as is shown in the figure.

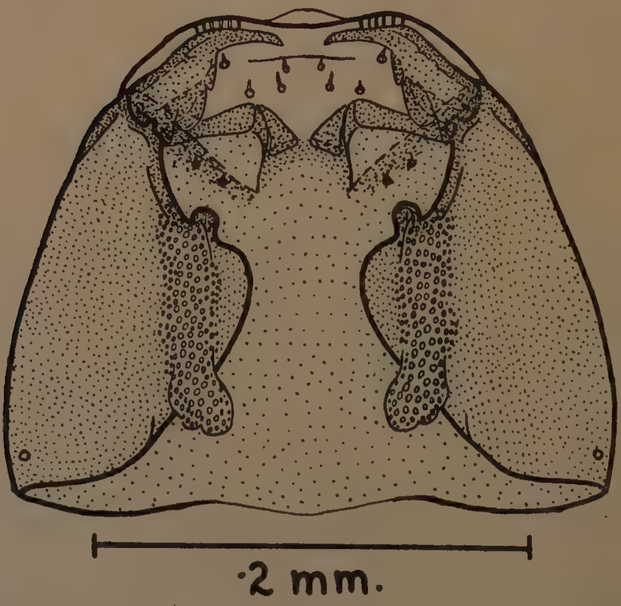

Fig. 3.—*Bracon vulgaris* (Cress.). First-stage larval head capsule.

The larva feeds on the body-fluid and possibly the fat-body of the host until in a few days the first instar larva is fully grown. At this time it is 2·5–2·8 mm. in length, and ·4–·5 mm. in width. The sides of the body are nearly parallel, and the "tail" has assumed a different shape from that when the larva had just hatched, being much smaller in relation to the size of the body. The abdominal appendages are slightly wider and blunter than on eclosion. The changes in both the "tail" and the abdominal appendages are those that would be expected from an increase in internal pressure within the parasite larva (fig. 4, *a*, *b*, *c*).

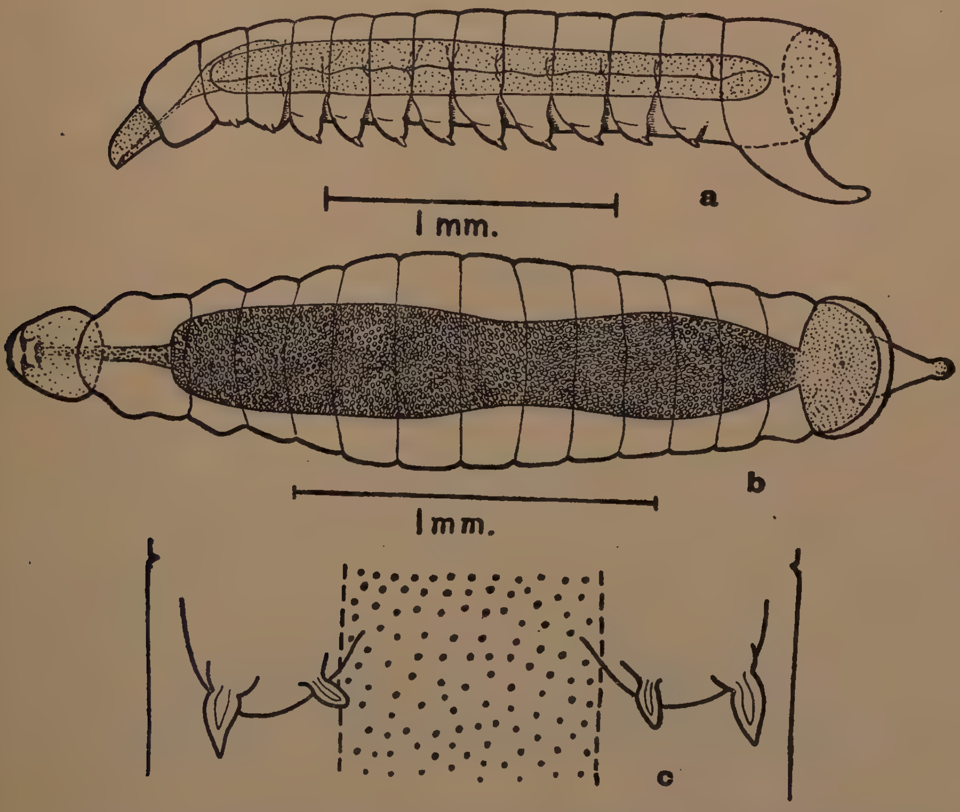

Fig. 4.—*Bracon vulgaris* (Cress.). Full-grown 1st-stage larva. (*a*) lateral view; (*b*) dorsal view; (*c*) ventral view of single abdominal segment, showing paired appendages.

The full-grown first-stage larva remains in the body cavity of the host until the latter has spun its cocoon, and undergoes no further growth until the physiological changes prior to pupation commence in the host. When the host larva enters a state of diapause in its cocoon instead of continuing its development, the *Bracon* larva is arrested at the full-grown first-stage, and no further change occurs until the state of diapause of the host comes to an end, when the development of the parasite continues as before. In this way complete correlation between the times of emergence of the generations of parasite and host is attained.

(*c*) *Second-stage Larva.*

This stage, intermediate in size, naturally, between the first and third stages, occurs at a period in the larval development when growth is very rapid. The larva is without outstanding distinctive characters. The head capsule and even the mandibles are without definite chitinisation and are sunk into the anterior body segments, the correct position of the sensoria being difficult to depict. The toothed mandibles are shown in fig. 5, and with the aid of these the full-grown second-stage larva bites to the exterior of the host. When it has done so, ecdysis occurs and the second stage larval skin remains attached to the host skin at the point of exit. The body of the larva is sac-like, has lost the paired segmental appendages, and the cauda is reduced to a vestige. The skin is ridged but without definite spines or papillae.

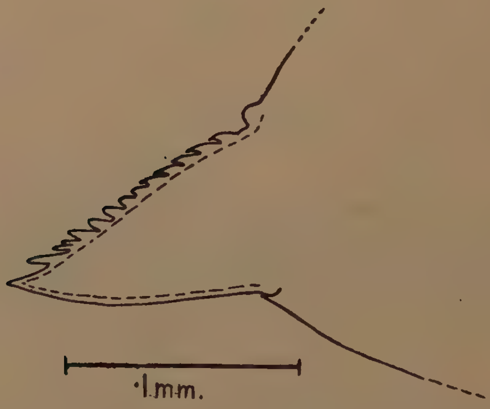

Fig. 5.—***Bracon vulgaris*** (Cress.). **Second-stage larva, mandible.**

The tracheal system is well developed, lateral trunks, anterior and posterior commissures and numerous segmental branches being visible ; segmental spiracular branches are present, but no spiracles were observed, and it seems that as in the first stage the tracheal system is without opening to the exterior. The larva in this stage begins to assume, by absorbing pigment from the host, the bright yellow colour seen in later stages.

(*d*) *Third-stage Larva.*

This is the final stage in the larval history and after ecdysis the early third-stage larva lies on the exterior of the host and finishes feeding almost ectoparasitically, sucking the hosts body contents through the hole made in the skin on emerging. The larva is then of the usual grub-like shape, though somewhat elongated, about ·8–1·0 cms. in length and about ·25 cms. wide.

The cephalic skeleton (fig. 6) is well developed, the mandibles are narrow and with fine teeth, the epistoma is indistinct, paired sensory setae are numerous, and there are two pairs of larger sensillae. The labial ring is well chitinised, and the opening of the silk duct distinct. The " tail " has become relatively much smaller, being now only a small projection at the posterior end, and the abdominal appendages have disappeared. The skin is somewhat ridged and is covered with small blunt papillae. The tracheal system is well developed and the spiracles are now functional (fig. 7).

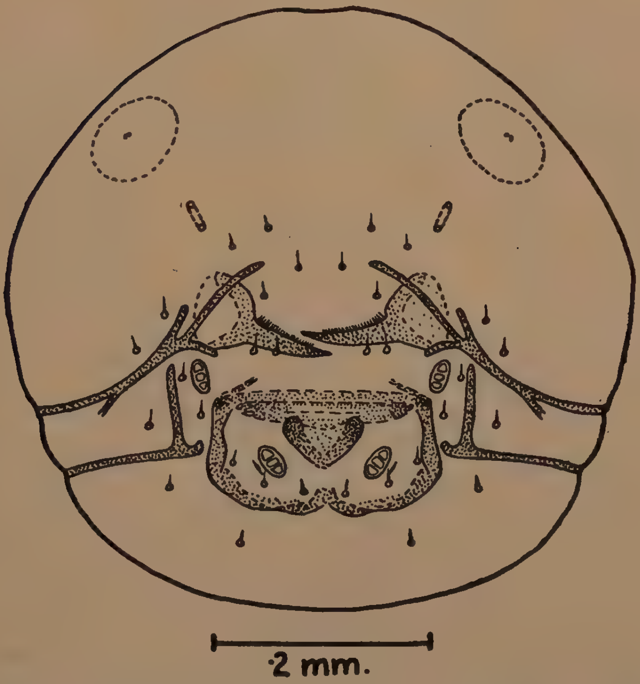

Fig. 6.—*Bracon vulgaris* (Cress.). Full-grown larvae, head capsule.

After feeding has been completed and the host has been entirely consumed, the parasite larva forms its cocoon within that of the host. The cocoon is relatively thin and flimsy, is made of fine silk, and has a shining appearance externally, with a thin white band around the middle. It is of the usual round-ended cylindrical shape and is 1–1·3 cms. in length, and ·3–·4 cm. in diameter.

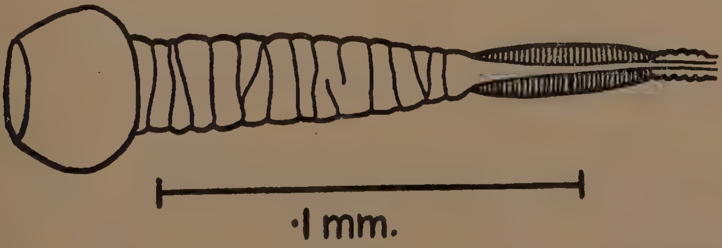

Fig. 7.—*Bracon vulgaris* (Cress.). Spiracle of full-grown larva.

Within the cocoon the larva forms a prepupa and pupa (fig. 8) from which the adult finally emerges. At 85°F. the egg stage lasts 5—6 days and, when development is uninterrupted, the first-stage larva is fully grown by about the end of the 7th–8th day. As stated above, when development occurs without diapause, the first stage larva remains as such while the host larva is growing, until cocoon formation occurs. The second-stage larva then lasts about one day, and the third stage for a further 1–2 days, when the cocoon is formed. There then elapses a further 10–12 days before emergence of the adult, of which 1–2 are passed in the prepupal stage.

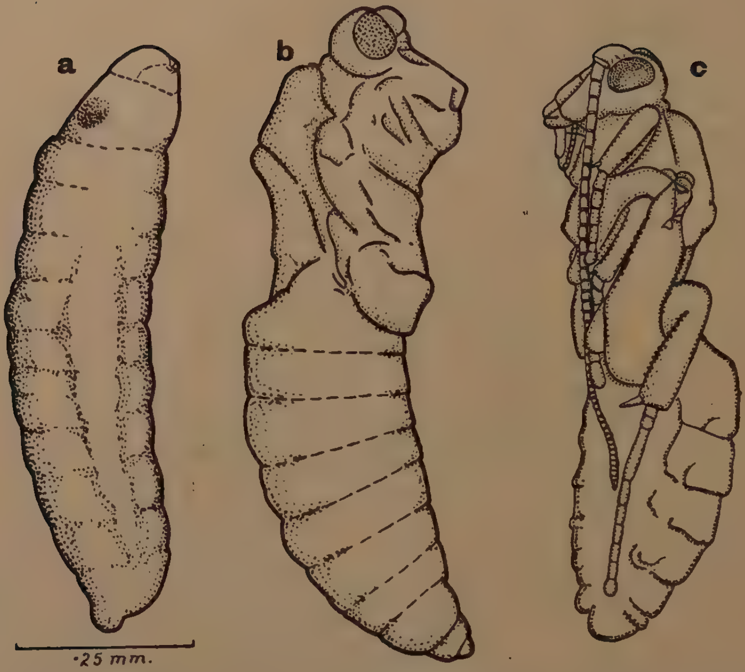

Fig. 8.—*Bracon vulgaris* (Cress.). (*a*) Prepupa, lateral view ; (*b*) Early pupa, lateral view ; (*c*) Fully developed male pupa, lateral view.

Importance.

From the figures given above it is obvious that *Bracon* is an important parasite of *Loxostege* and, although the percentage of hosts it destroys does vary considerably, it is usually the most abundant parasite reared from *Loxostege* material. The present work has dealt entirely with hibernating host material, but the other records indicate that *Bracon* is also important during the summer months in checking increase of the host. It is worth noting here that in September 1945, in Alberta, large numbers of adult *Bracon* together with other *Loxostege* parasites were observed on the wing in sugar-beet fields. The parasites flew from plant to plant and crawled all over the leaves obviously in search of hosts, sometimes as many as ten parasites being present on one plant. A number of moths (a partial second generation) had emerged at the same time as the parasites, but the weather was sufficiently cool to inhibit egg-laying and consequently no host larvae were present. Thus the synchronization of generations of host and parasites had in this case led to a definite waste of the latter, since these adults on the wing would inevitably die without producing progeny. This shows the advantage rather than the disadvantage of the biological control method, since had any *Loxostege* eggs been laid, parasites would have been already waiting to attack the resulting larvae, and the synchronization of generations thus ensures the emergence of parasites whenever host material is likely to be available. The species is apparently very specific in its host preferences, but it is hoped that on introduction into South Africa it will attack the Karoo caterpillar, *Loxostege frustalis*, Zell. It has been seen that the secondary parasite, *Mesochorus perniciosus*, Vier., destroys a varying percentage of *Bracon*, and possibly in South Africa, freed from the action of this secondary, the control exerted by *Bracon* on *Loxostege* will be considerable, if and when it becomes established.

The sporadic occurrence of *L. frustalis* and the specificity of the parasite host-relations will possibly diminish the effectiveness of parasitic control, but the same condition occurs with *L. sticticalis* in America where *Bracon* is able to maintain a fairly high level of parasitism. It is naturally impossible to predict whether this parasite will act as an effective controlling agent under the conditions occurring in South Africa, but this investigation of its biology indicates the possibilities of a successful introduction.

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THE OAT BIRD-CHERRY APHIS, *RHOPALOSIPHUM PADI*, L., AND COMPARISON WITH *R. CRATAEGELLUM*, THEO. (HEMIPTERA, APHIDIDAE).

By J. P. Rogerson, M.Sc. E.M.N
Department of Agriculture, King's College, Newcastle-upon-Tyne.

Introduction.

In Europe and America considerable confusion exists about the biology and morphology of certain species of *Rhopalosiphum* infesting grain. Börner (1931), pointed out that at least two closely allied species had been confused by other workers and included under the name Oat-Apple Aphis, *R. prunifoliae*, Fitch. Linnaeus (1767) gave the name *Aphis padi* to a green insect on *Prunus padus* that was later described by Koch. From Koch's description it is obvious that Linnaeus' species is the form typical for *Prunus padus*, which is a *Rhopalosiphum*, *i.e. R. padi*, L. Fitch (1855) gave the name *Aphis prunifoliae* to an American species with *Prunus* as the winter host and various Gramineae as the summer hosts. It now appears certain that he included more than one species under this name and that *R. padi* was one of them. Theobald (1927) in his " Aphididae of Great Britain " described and figured various stages of *R. prunifoliae*, Fitch, and in doing so confused two distinct species, *i.e. R. padi*, L., and another which Börner has stated should be called *R. crataegellum*, Theo. In the same work (Vol. II pp. 161–165) Theobald figured and described *Aphis avenae*, F., giving *Prunus padus* and *P. cerasus* as the winter hosts. Hille Ris Lambers (1934) pointed out that Theobald's species is *R. padi*, L. Theobald discussed the synonymy of Linnaeus' *padi* (Vol. II pp. 403–404) and assumed that it was a synonym of *Anuraphis helichrysi* despite the fact that he quoted from Koch's accurate description of *R. padi*, L.

In view of the economic importance and extraordinary confusion centering around the group, the author decided to investigate the biology and nomenclature of the grain-infesting species of *Rhopalosiphum*. An opportunity to commence a study arose in the summer of 1940 when a heavy infestation of grain aphids occurred on oats throughout the Northern Advisory Province. Since it was necessary to establish the identity of the species concerned, the present work is mainly related to the morphology and biology of *R. padi*, L., and *R. crataegellum*, Theo. The economic significance of these aphids has also been investigated and will be discussed later.

Rhopalosiphum padi, L.

Chief synonyms : *Aphis padi*, Linnaeus, Kaltenbach.
Aphis prunifoliae, Fitch (partim.).
Aphis avenae, Theobald, non Fabricius.
Siphocoryne avenae, Fab. (Theobald).
Siphocoryne splendens, Theo. (?)

The synonymy in American literature is very confused and the name *R. prunifoliae* is loosely used for both *R. padi*, L., and *R. annuae*, Oestlund.

Morphology.

Apterous viviparous female (fundatrix).

Colour : Body bright green, head slightly darker. Antennae pale green, apices of III and IV and whole of V and VI, dusky. Eyes dark red. Legs pale green, apices of femora dusky, apices of tibiae brown ; tarsi dark brown. Cornicles green, dusky at apices. Cauda pale at base, brownish on apical $\frac{2}{3}$. Abdomen with a rosy pink circular area at base of each cornicle. This area varies in size in different specimens; in some, it appears as a well-defined patch, in others, it may coalesce over the abdomen as one continuous irregular band.

Taxonomy of macerated specimen: Body ovate, without sclerotisation of the cuticle. Lateral papillae on pronotum and on abd. segs. 1–7, those on 7 being the largest. Small wax glands present on terga of meso- and metanota and on abd. segs. 1–5. Cuticle with fine reticulations. Antennae (fig. 1, *a*) very short, reaching to hind coxae, of 6 segments and without secondary rhinaria. Apex of III and whole of IV, V and VI, imbricated. Cornicles short, slightly vasiform, faintly imbricated, tips flared. Cauda about as long as cornicles, strongly spinulated, with many lateral curved spine-like setae. Genital plate entire, small. Anal and genital plates with fine spinules and many strong setae. Legs short, stout. Rostrum short, not reaching beyond second coxae.

Chaetotaxy: Head and body setae small and inconspicuous. Dorsal setae occur in a single row on each thoracic and abdominal segment. Ventral chaetotaxy much stronger and more profuse. Antennal hairs short and sparse. Legs with numerous hairs especially on the hind tibiae, where they are strong and spine-like.

Measurements: *vide* Table I.

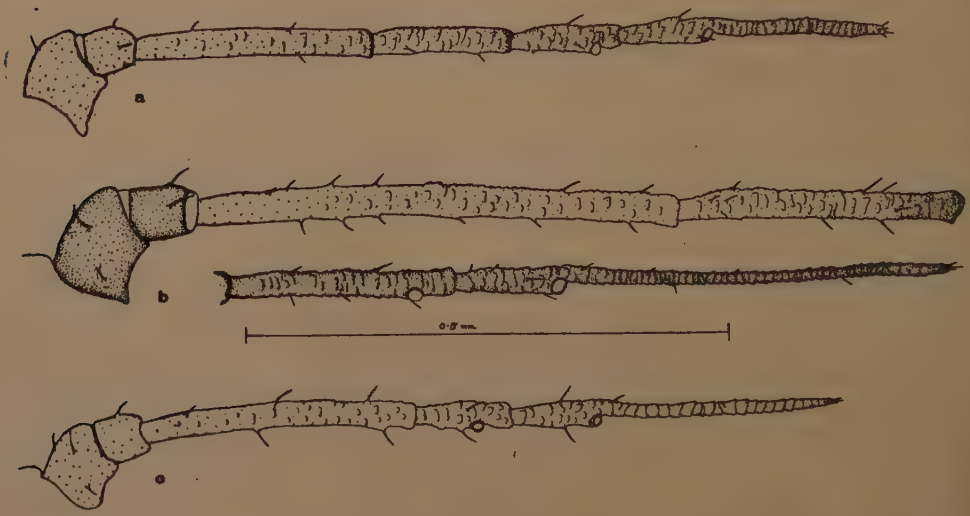

Fig. 1.—*Rhopalosiphum padi*, L. (*a*) Antenna of apt. viv. ♀ (fundatrix); (*b*) apt. viv. ♀ (fundatrigenia); (*c*) *R. crataegellum*, Theo., antenna of apt. viv. ♀ (fundatrix).

Apterous viviparous female (fundatrigenia).

Colour: Ground colour green, almost obscured by whitish flocculent meal which extends from pronotum down the lateral margins, the dorsal area being only thinly covered. Posterior segments from cornicles to cauda thickly covered. Rusty patch at base of cornicles visible indistinctly through the meal. Legs ochreous-green, apices of femora and tibiae dusky; tarsi dark. Antennae ochreous-green, apices of seg. IV and V and whole of VI dark. Cornicles and cauda dark. Venter green, not so mealy as dorsum.

Taxonomy of macerated specimen: Body ovate, without sclerotisation of cuticle. Wax glands which are larger than in fundatrix, present on terga of meso- and metanota and on abdominal segs. 1–5. Cuticle slightly rugose, with faint but well-defined reticulations. Antennae (fig. 1, *b*) shorter than body, of 6 segments, without secondary rhinaria. Apical $\frac{2}{3}$ of seg. III and whole of segs. IV, V and VI imbricated. Cornicles short, slightly swollen in middle, faintly imbricated and with a few apical striae; apices flared. Cauda shorter than cornicles, with numerous spinules and curved setae. Anal and genital plates with small spinules and a number of strong setae. Genital plate entire. Pronotal lateral papillae large, smaller lateral papillae on abdominal segs. 1–4 and on 7.

Chaetotaxy: Head with a seta on each frontal lobe; a seta on the vertex on each side of the dorso-median line between the insertion of the antennae; 2 setae posteriorly on each side of the dorso-median line between the eyes. Frons with 2 long setae between the frontal lobes and 4 shorter ones anterior to base of rostrum. Dorsally, pro- meso- and metanota and abdominal segs. 1–5 bear from 7–9 setae; abd. seg. 6 bears 5 setae, seg. 7 bears 3–4 and seg. 8 bears 2. These setae are borne in a single row across the seg. Ventrally, the setae are much more numerous and occur in a double row across each thoracic segment and on abd. segs. 1–6. The number on the abd. segs. is about 30. Antennae and legs with many hairs, those on hind tibiae being very numerous and spinelike.

Measurements: *vide* Table I.

Alate viviparous female (migrantes).

Colour: Head black. Thorax with meso- and metanotal lobes black, pronotum green. Antennae dark brown except extreme base of III, which is green. Eyes reddish. Legs ochreous green, apices of femora and tibiae dark, tarsi black. Cornicles dusky. Cauda green at base, dusky at apex. Abdomen green, slightly suffused with pink at base of cornicles; terga with dusky lateral patches. Venter pale green, genital and anal plates brown.

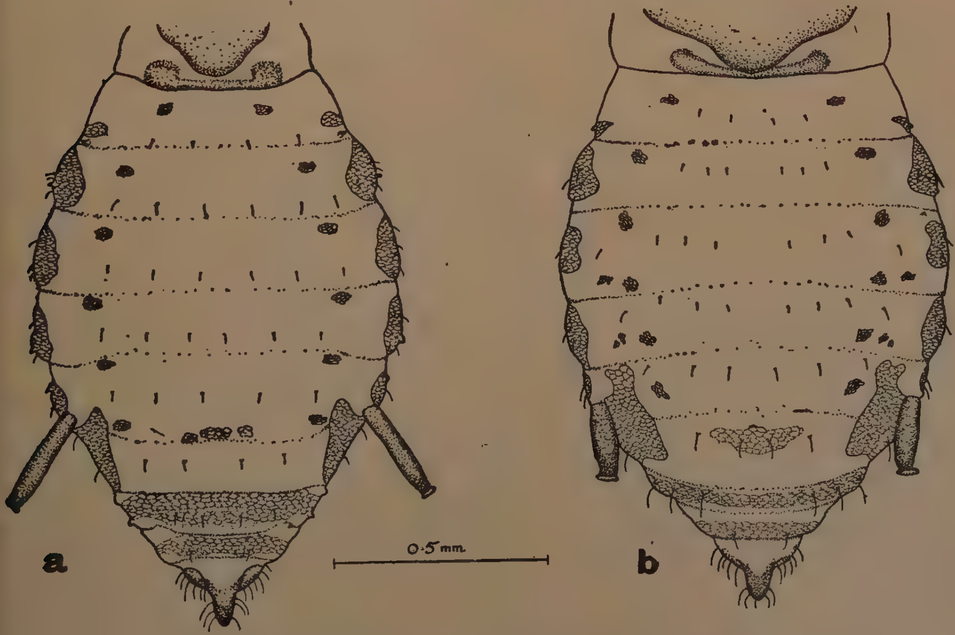

Fig. 2.—Body markings of al. viv. ♀ (migrantes). (*a*) *R. padi*, L.; (*b*) *R. crataegellum*, Theo.

Taxonomy of macerated specimen: Body oval, sclerotisation of cuticle as in living specimen. Head strongly chitinised, bearing prominent median and lateral stemmata. Frontal tubercles moderate. Eyes large. Antennae (fig. 4, *b*) reaching to near base of cornicles. Seg. I larger than II; III slightly imbricated, with 19–33 rhinaria along entire length; IV, V and VI more strongly imbricated, IV with 4–9 rhinaria, V with 0—2+1 rhinaria; VI with a primary rhinarium at base of flagellum; flagellum about 3¾ × length of base. Cornicles, cauda, anal and genital plates as in apterous viviparous female. Legs long and slender. Rostrum short, not reaching to second coxae. Abdominal terga with sclerotised plates laterally on segs. 1–6; segs. 7 and 8 each with a broken dorsal band. A small pleural tubercle is present on segs. 1–7 and wax glands on lateral margins of segs. 1–6. Lateral papillae present on pronotum, and on abd. segs. 1–4 and 7. Ventral cuticle with fine narrow reticulations.

Chaetotaxy: pleural, dorsal and ventral satee similar to those in apterous viviparous female, but those of head and thorax more numerous. Antennae with a number of short fine setae. Legs with many setae, those on femora and tibiae, especially of the hind legs, being strong and spine-like.

Measurements: *vide* Table I.

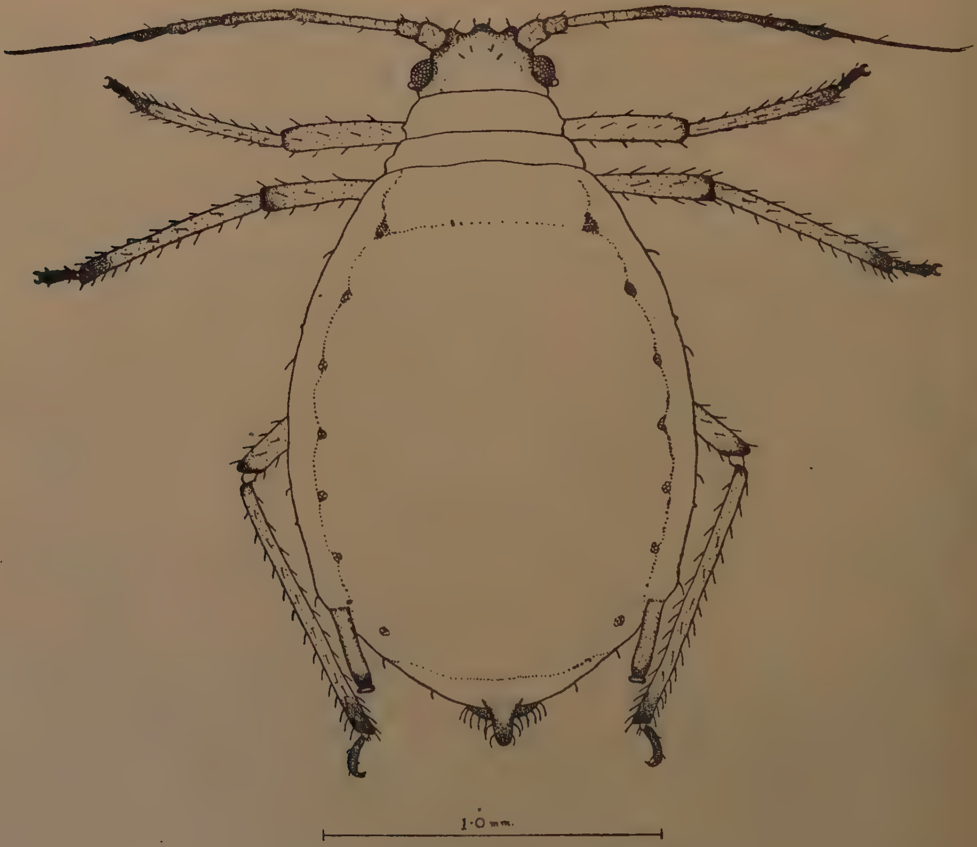

Fig. 3.—*R. padi*, L., apt. viv. ♀ (alienicola).

Alate viviparous female (alienicola).

Morphologically similar to the alate migrantes but smaller. The antennae are slender and bear fewer rhinaria than the other alate forms. In the structure of the lateral papillae it resembles the alate gynopara but the body chaetotaxy is like that of the alate migrantes. The 2 dorsal setae on abd. seg. 8 at most ⅔ width of ant. seg. III. Setae on III not more than ⅓ width of segment.

Measurements : *vide* Table I.

Apterous viviparous female (alienicola). Figure 3.

Colour : Dark green, abdomen with a rusty-pink area extending from base of cornicles to cauda. Eyes black. Antennal segments I, II and base of III green, rest brown. Legs green, apices of femora and tibiae brown ; tarsi black. Cornicles dusky green, dark at tips. Cauda green at base, dusky at apex. Rostrum green, tip dark. Wax glands on terga of meso- and metanota, and on abd. segs. 1–5, appear as small brown spots. Not mealy as in the fundatrigeniae.

Taxonomy of macerated specimen : Very similar to the apt. fundatrigenia but smaller. Frontal tubercles more strongly developed, antennae longer, cornicles and cauda longer and thinner. Tergal cuticle strongly reticulated. Lateral papillae small, inconspicuous, present on pronotum and on abdominal segs. 1, 3, 4 and 7.

Chaetotaxy : Dorsal setae shorter and sparser than in apt. fundatrigenia. The 2 dorsal setae on abd. seg. 8 not longer than width of ant. seg. III. Antennal setae on III not more than ½ width of the segment.

Measurements : *vide* Table I.

Alate viviparous female (gynopara).

Similar to the alate migrantes morphologically but smaller and darker. Flagellum of ant. seg. VI relatively longer, rhinaria on IV and V more numerous. Lateral papillae are very small and occur on pronotum and on abd. segs. 1 and 7.

Antennae *vide* fig. 4, *a*.

Measurements : *vide* Table I.

Alate male.

Colour : Darker than al. viv. female. Head, meso- and metanotal lobes black. Pronotum green. Antennae black. Eyes dark. Legs with base of femora and basal ⅔ of tibiae ochreous, rest black. Cornicles, cauda, anal and genital plates dark. Abdomen green, terga with dusky lateral and median markings. Venter green. Genital organs dark.

Taxonomy of macerated specimen : Body small, thorax large and strongly developed. Head strongly chitinised, with prominent median and lateral stemmata. Eyes prominent. Antennae (fig. 4, *d*) as long as the body ; seg. I larger than II, III imbricated, with 33–40 rhinaria along entire length ; IV, V and VI strongly imbricated, IV with 17–29 rhinaria, V with 11 − 18 + 1 rhinaria ; flagellum of VI 6 × length of base. Cornicles thinner and more cylindrical than in alate female, slightly swollen apically, imbricated. Cauda about ½ cornicles. Abdominal terga with sclerotised plates laterally on segs. 1–6 ; segs. 7 and 8 with a sclerotised transverse band : segs. 1–6 also bear median patches, which in some coalesce to form a single irregular line. Pleural tubercles and wax glands as in alate female. Abdominal cuticle with fine narrow reticulations on ventral surface. Pronotal lateral papillae small, none on abdomen.

Chaetotaxy : Similar to alate female.

Measurements : *vide* Table I.

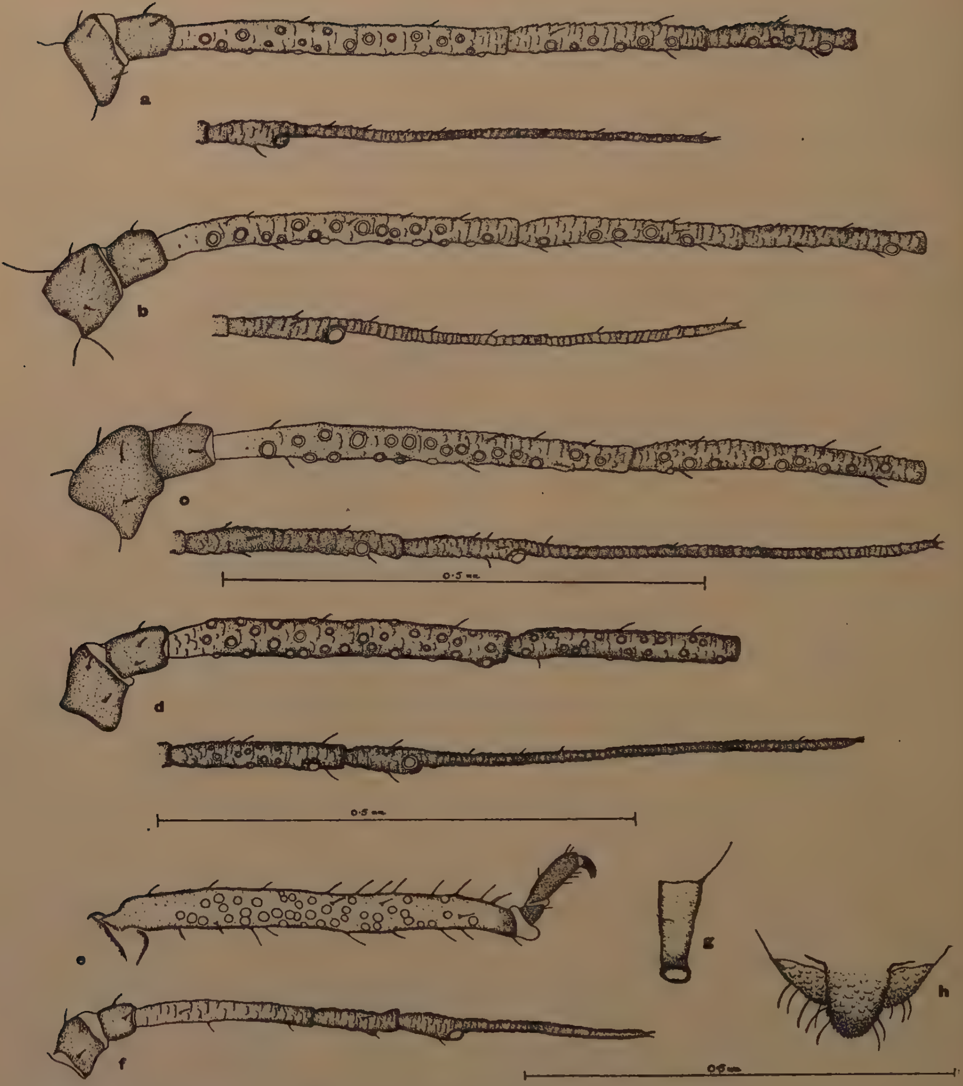

Fig. 4.—*R. padi*, L. Antenna of: (*a*) Alate viv. ♀ (gynopara); (*b*) Alate viv. ♀ (migrantes); (*c*) Alate viv. ♀ (progeny of fundatrigenia); (*d*) Alate ♂; (*e*) Hind tibia; (*f*) Antenna; (*g*) Cornicle; (*h*) Cauda; of apt. oviparous ♀.

Apterous oviparous female. Figure 5, *b*.

Colour: Green to yellowish-green, abdomen with lateral area and dorsal line darker. Eyes red. Antennae green with dusky apices. Legs dusky green, tarsi dark. Cornicles green, apices dark. Cauda green. Anal and genital plates dusky green.

Taxonomy of macerated specimen: Smaller and more elongated than apt. viviparous female. Antennae (fig. 4, *f*) short, of 5 segments, reaching just beyond hind coxae, without secondary rhinaria. Apex of seg. III and whole of IV and V imbricated. Cornicles (fig. 4, *g*) very short, broad at base and tapering towards apices, not swollen; imbricated, apices flared. Cauda (fig. 4, *h*) shorter than cornicles, spinulated, with lateral curved setae; rounded at apex. Genital and anal plates as in apterous viviparous female. Legs short, hind tibiae (fig. 4, *e*) broad, with 40–62 (mean 46·4) pseudosensoria on whole length. Rostrum short and stout, reaching just beyond second coxae. Cuticle slightly rugose, faintly reticulated. Pleural tubercles present on terga of meso- and metanota and on abd. segs. 1–7.

Chaetotaxy: Body hairs shorter and sparser than in apt. viviparous female.

Measurements: *vide* Table I.

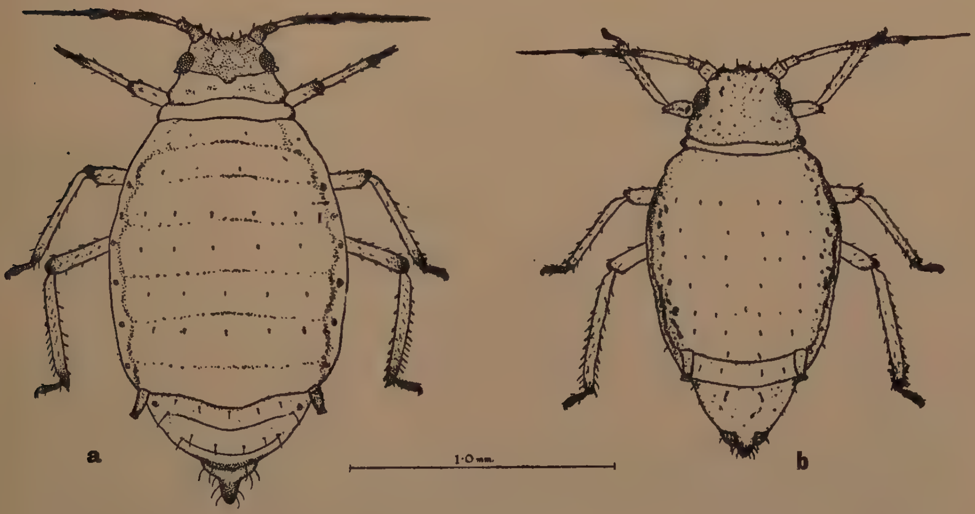

Fig. 5.—(*a*) Apt. oviparous ♀ of *R. crataegellum*, Theo.; (*b*) Apt. oviparous ♀ of *R. padi*, L.

Egg: Oval in shape. Green when laid, later turning shiny black. The ova are deposited in the bud axils, particularly on the young leading shoots. Counts of ova have shown as many as 38 per bud. Length 0·586 mm. width 0·261 mm. (mean measurements based on 20 ova).

Biology.

The eggs are laid by the oviparous females on bird-cherry in October and November, sometimes in September. These hatch out in April and the larval fundatrices commence feeding on the opening buds, often in such numbers as to cover the surface of them. The fundatrices collect on the undersides of the young leaves, generally along the midrib. They mature in about a fortnight and produce apterous young which remain together, so that each leaf bears a colony composed of the direct progeny of the resident fundatrices. The characteristic leaf-rolling commences after the production of fundatrigeniae and in no case was it observed until this generation developed. The leaf is rolled downwards parallel to the midrib, forming a cigar-shaped tube that protects the aphid colony. This rolling usually becomes evident about the time the flowers develop and, as the colonies increase in size, globules of honey-dew form inside the leave, which turn yellow and die. Alatae, the progeny of the fundatrigeniae, arise in May and with their appearance the colonies on bird-cherry begin to die out. By the end of June most of the alatae have migrated to corn and grasses, the foliage of the bird-cherry being by this time brown and sticky and covered

TABLE I.

Measurements of *R. padi* (in mm.)

(The arithmetic means are based on measurements of a large number of individuals from different colonies.)

Form	Body length	Antennal segments				Hind tibiae	Cornicles	Cauda	Antennal rhinaria		
		III	IV	V	VI				III	IV	V
Apt. viv. ♀ (fundatrix)	2·707	0·251	0·146	0·113	0·092+ 0·175	0·804	0·190	0·181	—	—	—
Apt. viv. ♀ (fundatrigenia)	2·730	0·463	0·285	0·240	0·117+ 0·416	1·143	0·280	0·178	—	—	—
Al. viv. ♀ (migrantes)	2·401	0·457	0·286	0·239	0·122+ 0·467	1·083	0·231	0·132	min. 19 max. 33 mean 23·6	4 9 7·2	0+1 2+1 0·47+1
Apt. viv. ♀ (alienicola)	1·907	0·297	0·172	0·144	0·080+ 0·389	1·011	0·234	0·138	—	—	—
Al. viv. ♀ (alienicola)	1·661	0·285	0·154	0·138	0·077+ 0·400	0·677	0·184	0·104	min. 12 max. 19 mean 14·3	2 7 3·8	0+1 1+1 0·31+1
Al. viv. ♀ (gynopara)	1·990	0·412	0·242	0·184	0·089+ 0·471	0·989	0·252	0·123	min. 16 max. 29 mean 22·6	6 11 8·1	0+1 6+1 1·84+1
Alate ♂	1·810	0·416	0·254	0·215	0·085+ 0·508	0·931	0·218	0·092	min. 33 max. 40 mean 37·1	17 29 22·8	11+1 18+1 14·3+1
Apt. ovip. ♀	1·507	0·224	0·098	0·062+ 0·231		0·449	0·108	0·077	—	—	—

with exuviae and mould. Rapid parthenogenetic reproduction takes place during summer and, under favourable conditions, the species becomes strongly established on grasses and cereals. On GRAMINEAE the aphids live in the leaf-sheaths and on the lower stems, seldom on the flower heads or grain. Towards autumn, the apterous generations decrease and alate females arise which fly back to *Prunus padus*, followed by the alate males. The oviparae are produced on bird-cherry by these return migrant females. Theobald suggests that it may overwinter as viviparae on GRAMINEAE, and in America Cutright (1925), Lathrop (1928) and Andre and Tate (1933) have recorded winter survival of *R. prunifoliae* (auct.) in this way. The latter workers found nymphs, alate and apterous viviparae overwintering in blue grass sods in Iowa, the mean temperature being 20°–50°F. and the monthly rainfall from ½–2 inches.

Food-plants.

(*a*) Winter hosts : *Prunus padus* in Britain and Europe. Börner gives *Prunus persicae* in Asia and *P. virginiana* in North America. Theobald states that it overwinters on *P. cerasus* but no evidence was obtained to support this view during the present investigation, although many hundreds of *cerasus* bushes were examined throughout the Northern Province. Even when growing next to heavily infested *P. padus* there was never any sign of colonies on the wild cherry. In Britain, at least, the species appears to overwinter only on bird-cherry.

(*b*) Summer hosts : various GRAMINEAE including oats, wheat, *Avena flavescens*, *Poa annua*, *Holcus lanatus*, *Agropyrum repens*, *Cynosurus cristatus*.

General.

Widely distributed throughout Europe and North America and probably also in other countries. It has been recorded from many parts of Britain and in some years becomes a serious pest of oats. During the summer of 1940 a severe outbreak occurred on oats in the Northern Province and considerable damage to crops resulted. A number of fields reported to be infested by "blight" or "fly" in Northumberland and Cumberland during June and July were examined and the damage attributed to this aphid. The aphids live in vast colonies on the lower stems and in the leaf-sheaths, checking and distorting growth by constant sucking of the plant juices. The most severe attacks occur on lea oats, especially on marginal land where cultural and climatic conditions are generally less favourable to rapid growth of the crop. Observations made during the years 1940–1944 indicated that the heaviest infestations developed in hill and semi-upland areas that prior to the war-time ploughing out of old grassland carried only a small tillage acreage. Thus, the areas most liable to serious attack are, in Northumberland, the North and South Tyne valleys, Coquetdale, the eastern slopes of the Pennines, and in Cumberland and Westmorland, the Lake District and the adjacent Pennine range. In these regions soil fertility and cultural conditions are not always adequate, and, normally, oats are sown later than is the case in the regular arable districts. In addition to these factors, weather conditions considerably influence the ability of the plants to withstand an attack, and at the same time affect the migration and colonisation of the aphids. During the warm dry spring of 1940 conditions for migration and rapid colonisation were good, with an absence of heavy rain that would have destroyed large numbers of migratory winged aphids. No serious attacks were observed in the Northern Province during the years 1941–1944 when the migratory periods were accompanied by high rainfall and low temperatures. In addition, the extent of re-infestation of the winter host influences the size of the colonies in the following year, and preliminary data indicates that the weather during the autumn migration has an important bearing on this re-infestation.

Overwintering of R. padi.

Special attention has been given to the overwintering of *R. padi* with a view to forecasting possible attacks during the following summer. In 1943 a method was devised of assessing the probable size of the spring colonies by estimating the number of viable ova on the winter hosts at certain centres. A random sample of 10 per cent. of the estimated total number of shoots per bush were removed and examined in the laboratory and the ova counted. Preliminary observations showed that the majority of these were laid on the first-year shoots and counts were therefore confined to the young wood. Counting the ova involved considerable time and labour since each ovum had to be examined under a low power binocular and its viability ascertained by pricking with a needle. Consequently, only four bushes were chosen for the purpose of ova counts, the sites of these being as follows :—

Site 1. Ulgham Oak Wood, Ulgham, Northumberland. This bush is a small one forming part of the hedge on the eastern edge of a mixed plantation. The first year shoots, estimated to number 550 for the whole bush, are rather spindly in growth. There are about 10 more bird-cherry bushes in the immediate vicinity.

Site 2. Low Streethouse, Calthwaite, Penrith, Cumberland. Here the bush forms part of a layered hedge which has not been cut for some years, so that the young wood is strongly grown. The total of first-year shoots on the bush is about 400. The site is exposed and there are no other bird-cherries near.

Site 3. Staward Peel, Whitfield, Northumberland. The two bushes A and B are located on the southern edge of a mixed plantation, in a well-sheltered position. Bush A, with an estimated total of 640 first-year shoots, occurs on the road-verge about five yards from the edge of the wood. Bush B, of 970 first-year shoots, lies just inside the plantation, 20 yards from A. There are a large number of *Prunus padus* bushes in the area of the site. The results of the first set of counts are given in Table II :—

TABLE II.

Counts of Ova on *Prunus padus.*

Bush ref.	Date sampled	No. of viable ova (on 10% shoots)	No. of non-viable ova (on 10% shoots)	Total no. of ova (on 10% shoots)	Percentage viable	No. of shoots on bush
Ulgham Oak Wood ...	27.1.44	949	121	1,070	88·69	550
Low Streethouse ...	22.2.44	1,201	401	1,602	74·97	400
Staward Peel A ...	3.3.44	3,069	1,050	4,119	74·51	640
Staward Peel B ...	3.3.44	442	91	533	82·93	970

It is interesting to note that there is a considerable difference in the number of ova on the Staward Peel bushes A and B, but that the degree of viability is fairly constant in all four bushes.

Further samples were taken from the same four bushes during the winter of 1944–45. In order to determine the possibility of a drop in the degree of viability, however, two lots of samples were taken from each bush, the first count of ova being made immediately after oviposition in November and after the ova turned black, and the second count in February before hatching. The results of these counts are tabulated below :—

TABLE III.

Counts of Ova on *Prunus padus.*

Bush ref.	Date sampled	No. of viable ova (on 10% shoots)	No. of non-viable ova (on 10% shoots)	Total no. of ova (on 10% shoots)	Percentage viable	No. of shoots on bush
Ulgham Oak Wood ...	13.11.44	183	62	245	74·69	550
Low Streethouse ...	23.11.44	565	290	855	66·08	400
Staward Peel A ...	22.12.44	1,104	576	1,680	65·71	640
Staward Peel B* ...	22.12.44	78	33	111	70·27	970

*Only 5 per cent. of shoots taken.

Comparison of Tables II and III shows that in each case the number of ova present on the corresponding bushes was much less during the 1944–45 overwintering period than in the year before. This may be explained by the fact that the wet, cold summer of 1944 hampered rapid reproduction of the aphids and that the consequent return migration to *Prunus padus* in the autumn was sparse. Also, it is probable that large numbers of winged return migrants were destroyed by heavy rain which fell during the migratory period of September–October.

TABLE IV.

Counts of Ova on *Prunus padus.*

Bush ref.	Date sampled	No. of viable ova (on 10%) shoots)	No. of non-viable ova (on 10% shoots)	Total no. of ova (on 10% shoots)	Percentage viable	No. of shoots on bush
Ulgham Oak Wood ...	13.2.45	46	114	160	28·75	550
Low Streethouse ...	21.2.45	165	187	352	46·87	400
Staward Peel A ...	22.2.45	984	433	1,417	62·38	640
Staward Peel B ...	22.2.45	129	92	221	58·37	970

As might be expected, Table IV shows a reduction in viability on every bush as compared to Table III. This reduction may be largely due to climatic conditions, but other factors, such as the presence of parasites and predators, may also be expected to affect the degree of viability, In addition, it will be seen from these tables that the total number of ova on each bush after the second lot of counts (Table IV) is lower than after the first counts (Table III), with the exception of Bush B, Staward Peel, from which only 5 per cent. of the shoots were examined in December 1944. In order to determine the margin of error involved in the random sampling of 10 per cent. of the bushes, the standard error (S.E.) has been calculated for the Ulgham site. Two 10 per cent. samples, totalling 110 shoots, had previously been removed from this bush, the first on 13.11.44 and the second on 13.2.45. The remaining eight 10 per cent. samples, totalling 440 shoots, were removed on 23.2.45 and the number of ova present on them counted in the laboratory. Counts of ova on these eight samples gave the following figures :—

TABLE V.

Sample No.	1	2	3	4	5	6	7	8
No. of ova	166	174	183	125	129	139	167	143

Mean $(\bar{x}) = 153 \cdot 25$ S.E. $= \pm 21 \cdot 915$

Distribution of Prunus padus *in the Northern Province.*

One of the factors affecting the incidence of *R. padi* as a pest of oats lies in the distribution of the winter host. Bentham and Hooker, in their " Handbook of the British Flora ", give the geographical areas of distribution as northern and central Europe and Asia but absent from S.W. Europe, southern England and a great part of Ireland. In order to gain a more precise knowledge of the distribution in the Northern Province, a survey of bird-cherry sites was commenced during the winter of 1943–44. Preliminary data indicates that the tree is a semi-upland species and is most numerous in the hill areas of the four northern counties. In Cumberland and Westmorland the tree is very common, especially in the Lake District, the Pennine foothills, including Vale of Eden, the area of Brough, Appleby and Kendal, Underbarrow and the Kent Valley. It is not so common on the western seaboard of Cumberland but occurs towards the heads of Eskdale, Wasdale, Ennerdale and Duddon Valley. The Northumberland sites are more or less localised to that part of the county west of an arbitrary line drawn through Wooler Alnwick and Hexham, but single trees and bushes are found even in the flat coastal plain. The chief sites in Northumberland are the valleys of the North and South Tyne rivers, Coquetdale, Glendale and the Cheviot foothills. Records for Durham are incomplete but numerous sites occur in Weardale, and in the Derwent Valley. The bushes often form an important part of farm hedges and are found in open woods, sometimes as a semi-ornamental tree in parks and private grounds.

In general it will be noticed that the areas of distribution correspond closely to the localities in which infestations of *R. padi* are most severe.

Rhopalosiphum crataegellum, Theobald.

Chief synonyms : *Aphis crataegi*, Buckton.
Rhopalosiphum prunifoliae, partim. (Theobald, etc.).

According to Börner (1931) the European " Apple-Grain Aphid ", *R. crataegellum*, Theo., is distinct from the American species which he says is *R. annuae*, Oestlund. If this is so, then much of the American literature dealing with *R. prunifoliae*, Fitch, is in fact referring to Oestlund's species.

Morphology.

Apterous viviparous female (fundatrix).

Colour : Bright green, abdomen with deep green dorsal line and lateral margins. Eyes dark. Antennae pale green, apices of IV and whole of V dusky. Legs green, apices of tibiae and all tarsi, dusky. Cornicles yellowish green, tips dark. Cauda, anal and genital plates dusky green.

Taxonomy of macerated specimen : Body ovate, without sclerotisation of cuticle. Lateral papillae present on pronotum and on abdominal segs. 1–7, those on pronotum and seg. 7 being larger than the others. Pleural tubercles present on terga of meso- and metanota and on abdominal segs. 1–7. Ventral cuticle with fine transverse reticulations. Antennae (fig. 1, *c*) short, reaching to about hind coxae, of 5 segments, without secondary rhinaria. Apex of III and whole of IV and V imbricated.

Cornicles of moderate length, slightly vasiform on apical half, faintly imbricated, tips flared. Cauda bluntly lanceolate, distinctly shorter than cornicles, with lateral curved setae and strongly spinulated. Anal and genital plates with fine spinules and long curved setae. Legs short, rather slender.

Chaetotaxy : Head and body setae long. Dorsal setae occur in a single row on each thoracic and abdominal segment. Ventral setae stronger, more profuse and occur in an incomplete double row on each segment. Chaetotaxy of appendages normal.

Measurements : *vide* Table VI.

The fundatrix of this species is very distinct from that of *R. padi*, especially when alive. The body is brighter green, more shining, and never has the rusty coloration at the base of the cornicles which is so characteristic of *padi*. It also differs in having only 5 jointed antennae, more slender legs and a stronger body chaetotaxy.

Alate viviparous female (fundatrigenia).

Colour : Head and thoracic lobes black and shining. Abdomen green, shining, with dark pigmented areas laterally and 2 transverse bars between cornicles and cauda. Antennae dark, extreme base of III paler. Legs green with most of femora, apices of tibiae and whole of tarsi dark. Cornicles dark. Cauda dusky green. Venter pale green. Anal and genital plates dark.

Taxonomy of macerated specimen : Head strongly chitinised, with prominent median and lateral stemmata. Frontal tubercles moderate. Eyes large. Antennae (fig. 7, *c*) reaching beyond third coxae. Seg. 1 larger than II, III slightly imbricated, with 13–19 rhinaria along entire length, IV, V and VI strongly imbricated, IV with 6–11 rhinaria, V with 0–2+1 primary rhinarium, VI with a large primary rhinarium at base of flagellum ; flagellum about 4¼ × length of base. Cornicles rather short, vasiform on apical half, strongly imbricated on unswollen basal half, more faintly on remainder, tips flared. Cauda anal and genital plates spinulated, with long curved hairs. Legs long and slender. Rostrum reaching to about second coxae. Abdominal terga with sclerotised lateral plates on segments 1–6, seg. 7 with a broken transverse bar, seg. 8 with a more or less complete transverse bar. Lateral papillae present on pronotum and on abdominal segments 1–7, and wax glands laterally on segments 1–6. Ventral cuticle with narrow fine reticulations. Pleural tubercles are present on abdominal segments 1–7.

Chaetotaxy : Head, antennae and thoracic lobes with short straight setae. Dorsal abdominal setae occur in a single row across each segment, those on the segments caudad of cornicles being longest. Ventral abdominal setae more profuse, especially on seg. 6. The ventral setae are most numerous along the median line, except on seg. 6 where they extend to the lateral margins. Cauda, anal and genital plates with many long curved hairs. Legs with short spinelike setae especially on the tibiae.

Measurements : *vide* Table VI.

Alate viviparous female (migrantes). Fig. 2, *b*.

The migrantes differs only slightly from the alate fundatrigenia. It is generally rather smaller and the rhinaria on antennal segments III and V are more numerous (fig. 7, *b*).

Measurements : *vide* Table VI.

Apterous viviparous female (alienicola).

Colour : Yellowish green, head ochreous. Antennae ochreous, apices of III and whole of IV and V dusky. Legs dusky, base of femora and tibiae ochreous. Cornicles dark. Cauda green.

Taxonomy of macerated specimen: Body ovate, without sclerotisation of cuticle. Lateral papillae present on pronotum and on abdominal segments 1–7, those on pronotum and on seg. 7 being largest. Wax glands absent. Dorsal cuticle transversely reticulated. Ventral cuticle narrowly striated. Antennae (fig. 6, *a*) short, reaching just beyond second coxae, of 5 segments, without secondary rhinaria. Cornicles short and thick, slightly vasiform, imbricated, tips strongly flared. Cauda, anal and genital plates similar to fundatrix. Legs short and stout.

Chaetotaxy: Similar to that of fundatrix but the setae are longer and more spinelike. Abdominal seg. 8 with 2 very long dorsal setae, at least 1½ × width of antennal seg. III. Antennal setae long, the longest on III being at least equal to the width of the segment.

Measurements: *vide* Table VI.

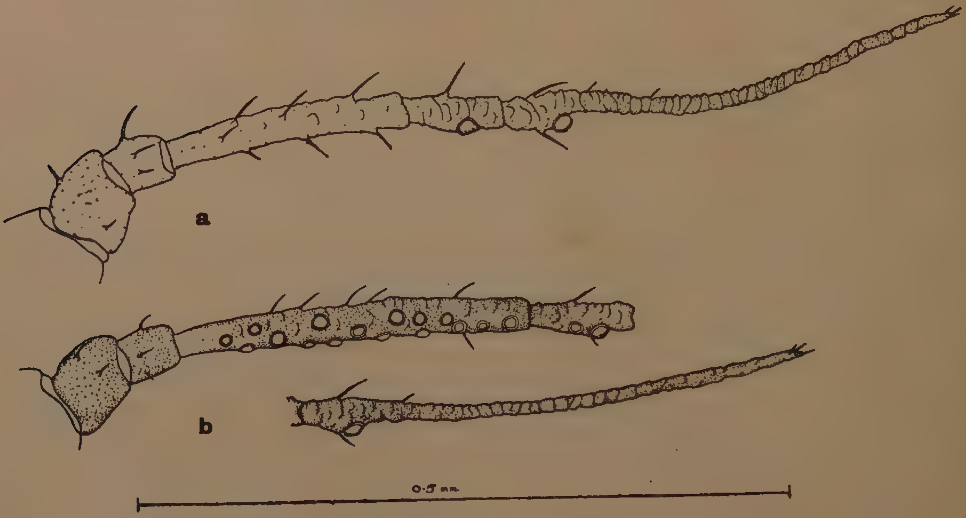

Fig. 6.—*R. crataegellum*, Theo. Antenna of: (*a*) Apt. viv. ♀ (alienicola); (*b*) Alate viv. ♀ (alienicola).

Alate viviparous female (alienicola).

Colour: Similar to alate migrantes.

Taxonomy of macerated specimen: Resembles the alate fundatrigenia. It differs from all the other alate forms in having only 5-segmented antennae (fig. 6, *b*). Segment III with 12–19 rhinaria, IV with 0–2+1 primary rhinarium; base of V with a large primary rhinarium; flagellum more than 5 × length of base. Abdominal seg. 8 with 2 long dorsal setae which are at least 1¼–1½ × width of antennal seg. III. Antennal setae on III at least ½ width of the segment.

Measurements: *vide* Table VI.

Alate viviparous female (gynopara).

Very similar to the alate fundatrigenia but with larger sclerotised areas on the abdomen. The antennae (fig. 7, *a*) are thicker, especially the base of segment VI, compared to the spring alatae. Lateral papillae are very small and occur on the pronotum and on abdominal seg. 7, rarely on any other segment.

Measurements: *vide* Table VI.

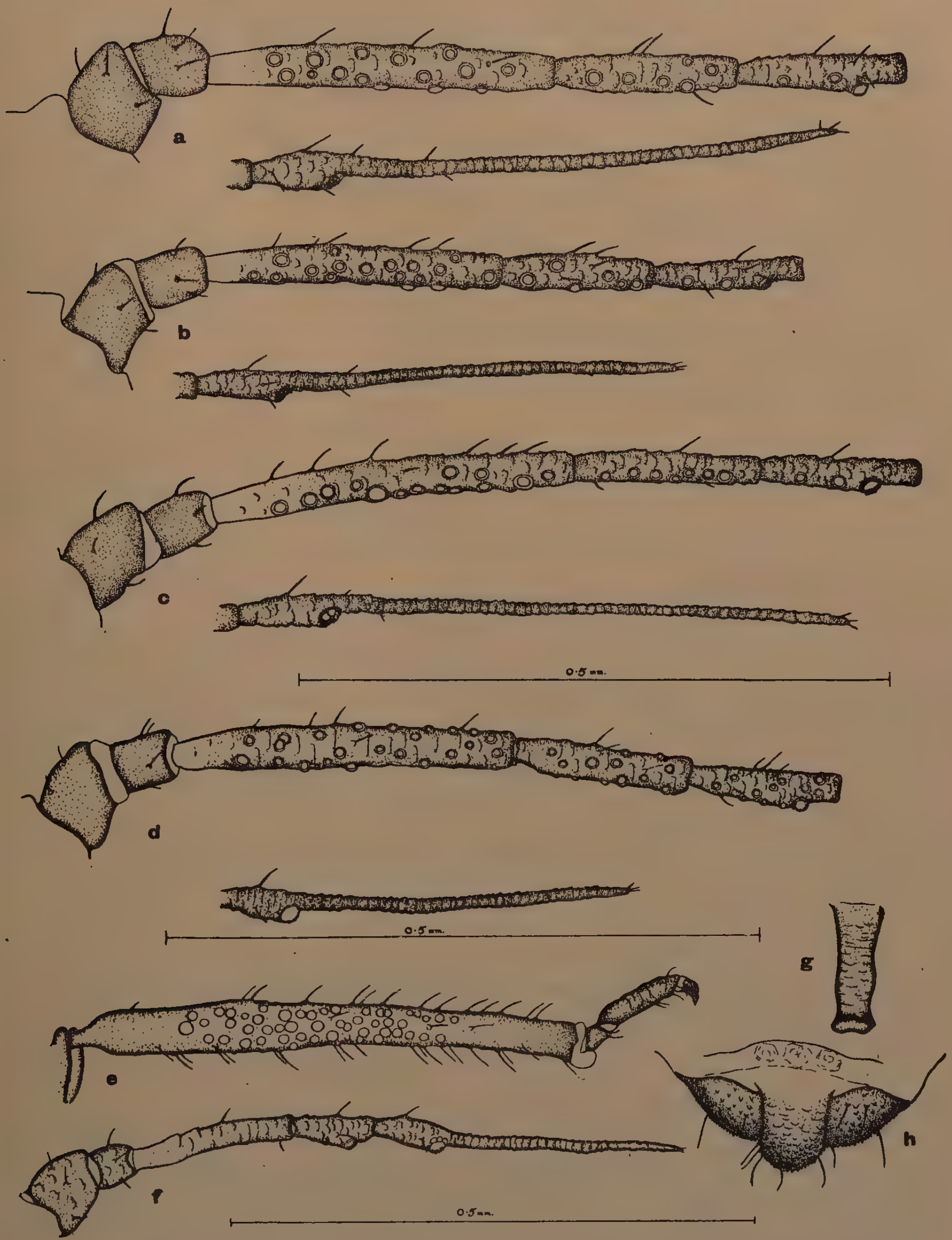

Fig. 7.—*R. crataegellum*, Theo. Antenna of: (*a*) Alate viv. ♀ (gynopara); (*b*) Alate viv. ♀ (migrantes); (*c*) Alate viv. ♀ (fundatrigenia); (*d*) Alate ♂; (*e*) Hind tibia; (*f*) Antenna; (*g*) Cornicle; (*h*) Cauda of apt. oviparous ♀.

Alate male.

Colour: Head and thoracic lobes black and shining. Antennae dark except small area at base of III. Eyes reddish. Legs ochreous, most of femora, apical $\frac{1}{5}$ of tibiae and all tarsi, black. Cornicles black. Cauda, anal and genital plates dusky. Genitalia dark. Abdomen green, terga with dark median and lateral markings.

Taxonomy of macerated specimen : Body small, thorax large and well-developed. Head with prominent median and lateral stemmata. Eyes large. Antennae (fig. 7, *d*) shorter than body, reaching to about 4th abdominal segment, imbricated, seg. I larger than II, III with 21–27 rhinaria, IV with 8–17 rhinaria, V with 8–13 rhinaria, sometimes a secondary rhinarium occurs on VI in addition to the normal primary one ; flagellum of VI, 5× length of base. Cornicles shorter than in alate female, slightly swollen apically, imbricated. Cauda small, ½ cornicles. Abdominal terga with lateral and median sclerotised plates on segs. 1–6. These coalesce to form a single transverse band on segs. 7 and 8. Pleural tubercles and wax glands as in alate female. Ventral abdominal cuticle with narrow fine reticulations. Pronotal lateral papillae small, none on abdomen.

Chaetotaxy : Similar to alate female.

Measurements : *vide* Table VI.

Apterous oviparous female (fig. 5, *a*).

Colour : Yellow to olive green with sclerotised brown areas on head. Antennae dusky green, base of III pale. Legs ochreous, tips of femora apices of tibiae and all tarsi dusky. Cornicles, cauda, anal and genital plates dusky. Presence of ova gives the abdomen a mottled appearance.

Taxonomy of macerated specimen : Ovate, smaller than apterous viviparous female. Antennae (fig. 7, *f*) short, scarcely reaching to third coxae, of 5 segments without secondary rhinaria, segments III to V imbricated. Cornicles (fig. 7, *g*) very short, slightly vasiform, imbricated, apices flared. Cauda (fig. 7, *h*) shorter than cornicles, spinulated, with strong curved lateral setae, rounded apically. Anal and genital plates as in apterous viviparous female. Legs short and stout, hind tibiae (fig. 7, *e*) slightly swollen with 45–63 (mean 54·7) pseudosensoria on middle part. Rostrum reaching beyond second coxae. Ventral cuticle faintly reticulated. Pleural tubercles present on meso- and metanota and on abdominal segs. 1–6.

Chaetotaxy : Body hairs much as in apterous viviparous female but shorter and sparser. Tergum of abdominal seg. 8 with 2 median setae that are longer than the other body hairs.

Measurements : *vide* Table VI.

Egg : Oval in shape, green when laid, later turning shiny black. The ova are deposited in cracks in the bark of the older shoots, and in the axils of the spurs. The smooth terminal shoots are not favoured for oviposition probably because they afford little protection against the weather.

Biology.

The general biology is similar to that of *R. padi*. Eggs are laid on apple, pear, rowan and hawthorn, more rarely on the other winter hosts, during September to November, most in October. They hatch in April, the date depending on the weather, and the fundatrices live in the unfolding buds. When the leaves develop the fundatrices and their progeny feed on the underside and form a loose type of leaf-curl that differs from that caused by *R. padi* in being rolled from tip to base and not edge to edge. Leaves so rolled do not change colour. Where colonies of *R. crataegellum* and the Rosy Aphis (*Yezabura malifoliae*, Fitch) are intermixed under the same leaf then discoloration occurs but is caused by *Yezabura*. The form and number of generations produced by the fundatrices varies, but observations made in the Northern Province during 1944–45 showed that only alate fundatrigeniae arose (*cf. R. padi*). Alate migrantes arise in May–June and fly away to found the alienicolae generations which live on GRAMINEAE during summer. The alienicolae are subterranean in habit and

TABLE VI.

Measurements of *R. crataegellum* (in mm.).

(The arithmetic means are based on measurements of a large number of individuals from different colonies.)

Form	Body length	Antennal segments				Hind tibiae	Cornicles	Cauda	Antennal rhinaria		
		III	IV	V	VI				III	IV	V
Apt. viv. ♀ (fundatrix)	2·341	0·302	0·101	0·088+ 0·261		0·846	0·227	0·159	—	—	—
Al. viv. ♀ (fundatrigenia)	2·132	0·317	0·193	0·162	0·091+ 0·387	0·963	0·215	0·113	min. 13 max. 19 mean 16·3	6 11 9·5	0+1 2+1 0·93+1
Al. viv. ♀ (migrantes)	1·815	0·278	0·139	0·144	0·073+ 0·375	0·795	0·168	0·108	min. 17 max. 21 mean 18·6	6 10 7·8	1+1 5+1 2·5+1
Apt. viv. ♀ (alienicola)	1·646	0·191	0·077	0·061+ 0·297		0·598	0·189	0·101	—	—	—
Al. viv. ♀ (alienicola)	1·421	0·275	0·089	0·062+ 0·344		0·617	0·154	0·089	min. 12 max. 19 mean 16·5	0+1 2+1 1·05+1	
Al. viv. ♀ (gynopara) ...	1·958	0·277	0·138	0·129	0·073+ 0·399	0·832	0·190	0·103	min. 14 max. 21 mean 17	4 9 7	0+1 4+1 2+1
Alate ♂	1·799	0·279	0·146	0·141	0·064+ 0·362	0·762	0·146	0·069	min. 21 max. 27 mean 24·5	8 17 13·7	8+1 13+1 9·9+1
Apt. ovip. ♀	1·400	0·154	0·077	0·062+ 0·239		0·457	0·108	0·077	—	—	—

feed on the roots and underground stems of grasses, the alatae moving to the aerial parts only as a prelude to migration to a fresh plant. The alate viviparae (gynoparae) and males return to the winter host in autumn, the former producing the oviparous females on these hosts in September and October.

Food-plants :

(*a*) Winter hosts : *Pyrus Malus* (wild crab and cultivated varieties), *Pyrus communis*, *Pyrus Aucuparia*, *Crataegus Oxyacantha*, *Pyrus Aria*, *Pyrus torminalis*, *Cotoneaster frigida*, *Cotoneaster pannosa*. Has also been recorded on *Mespilus* and *Pyrus Cydonia*. In Britain, the apple is the chief winter host.

(*b*) Summer hosts : various GRAMINEAE, including *Poa annua*.

General.

Very widespread throughout Britain and probably also in Europe. In Britain, however, it does not appear to assume the importance of a cereal pest to the extent of *R. padi*. As an apple pest it is one of three species responsible for considerable loss of crop annually, particularly in orchards that are not regularly sprayed. The constant sucking of the sap in the young growths and developing blossom trusses checks the free formation of fruit and sometimes results in the premature fall of fruitlets. Control measures lie in spraying the trees with an ovicidal wash in winter, or alternatively using a nicotine or similar contact insecticide just after the eggs hatch.

In America, Sullivan and McBride (1924) record that *Siphonaphis* (*Rhopalosiphum*) *padi* was the worst aphid pest on apple in Missouri in 1923–24. Lathrop (1928), on the other hand, dealing with the biology of the apple aphids, *Anuraphis roseus* (*Yezabura malifoliae*), *Aphis pomi* and *R. prunifoliae* in Ohio, says that the latter species has little effect on the fruit compared with the other two. Owing to the state of the American nomenclature on this group it is impossible, without examining typeslides, to decide whether these workers are discussing *annuae* or *crataegellum*.

Drake, Tate and Harris (1932), working on the transmission of the virus disease " yellow dwarf " of onions in Iowa, found that *R. prunifoliae* was one of the vectors of the disease. So far, no evidence has been forthcoming to suggest that *padi* or *crataegellum* occur on onions in Britain.

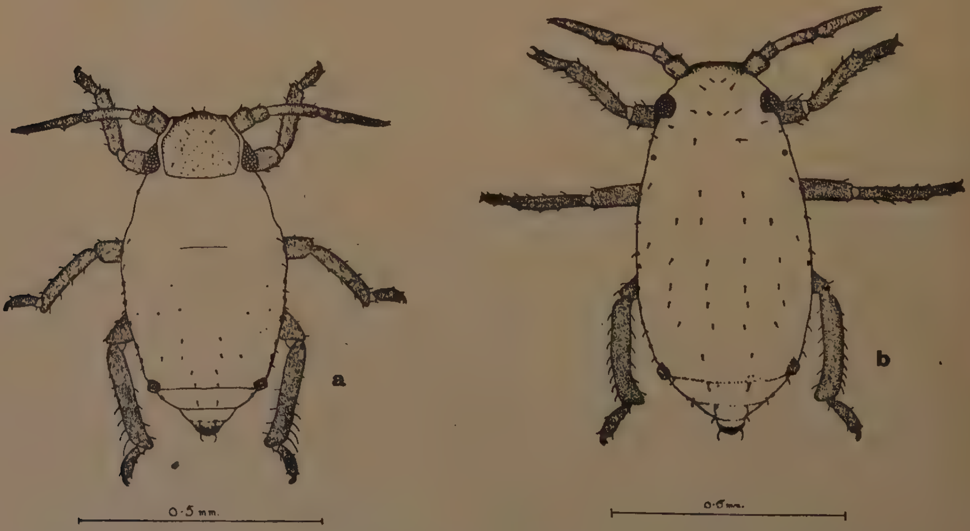

Fig. 8.—First-instar larval fundatrix of : (*a*) *R. padi*, L. ; (*b*) *R. crataegellum*, Theo.

Comparison of R. padi and R. crataegellum.

The two species have already been described in detail, so that to avoid unnecessary repetition only the chief distinguishing characters will be given here.

1. In *padi* the spring and summer forms show a characteristic rusty-red coloration on the abdominal tergum in the vicinity of the cornicles ; *crataegellum* is never ornamented in this way. The difference is more marked in the apterous than in the alate forms.

2. The fundatrigeniae of *padi* are covered with white waxy meal, and the fundatrices are also slightly mealy. None of the stages of *crataegellum* exhibit any marked degree of farinosity.

3. The fundatrix of *padi* has 6 antennal segments ; *crataegellum* only 5.

4. In *padi* the first fundatrigenia generation is apterous ; in *crataegellum* it is alate.

5. The alienicolae generations of *padi* and *crataegellum* differ considerably both in structure and in habits. In *padi*, the alate and apterous alienicolae have 6 segmented antennae ; in *crataegellum* only 5 antennal segments occur. The alienicolae of *padi* live on the aerial parts of GRAMINEAE, those of *crataegellum* live on the roots, the alatae only moving above soil level prior to dispersal to a fresh plant.

6. The alatae of *padi* have more antennal rhinaria than the corresponding forms of *crataegellum*. This is particularly marked in the case of the males.

7. Pseudosensoria on the hind tibiae of the oviparous female of *padi* extend over the whole length ; in *crataegellum* they occur on the middle of the tibiae.

8. Larval fundatrices of *padi* are dark green when newly hatched, paling after the first two or three moults. Larvae of *crataegellum*, on the other hand, are pale green and differ little from the adult fundatrix in colour.

9. In Britain, the winter host of *R. padi* is *Prunus padus*. *R. crataegellum* overwinters on various ROSACEAE, particularly *Pyrus* spp. but never on *Prunus* species.

Summary.

1. An attempt has been made to elucidate the confusion existing about the biology, morphology and nomenclature of the Oat-Apple Aphis, *Rhopalosiphum prunifoliae*, Fitch. A close study of the problem confirms the observations made by Börner that at least two distinct species have in the past been included under the name of *R. prunifoliae*. The names *Rhopalosiphum padi*, L., and *Rhopalosiphum crataegellum*, Theo., are adopted for these species.

2. Detailed descriptions and figures of the different generations are given and a comparison shows that the two species can readily be distinguished from one another.

3. *R. padi* appears to be of greater importance as a cereal pest than *R. crataegellum*. The latter species is a widespread pest of pip-fruit.

4. An account is given of the overwintering of *R. padi* on *Prunus padus*, and the results of experiments on estimating the size of the ova-populations is shown.

The author is indebted to Mr. R. A. Harper Gray for allowing him facilities to carry out the investigation ; also to Mr. J. A. Dunn for assistance in collecting some of the material and much general help.

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THE MALARIA PROBLEM IN MAURITIUS:
THE BIONOMICS OF MAURITIAN ANOPHELINES.

By W. F. Jepson, Ph.D. (Lond.),
Entomologist, Department of Agriculture, Mauritius,

A. Moutia, Dip. Agr. (Maur.),
Assistant Entomologist, Department of Agriculture, Mauritius,

and

C. Courtois, Dip. Agr. (Maur.),
Technical Assistant, Malaria Control Unit, Mauritius.

abstr. L.C.

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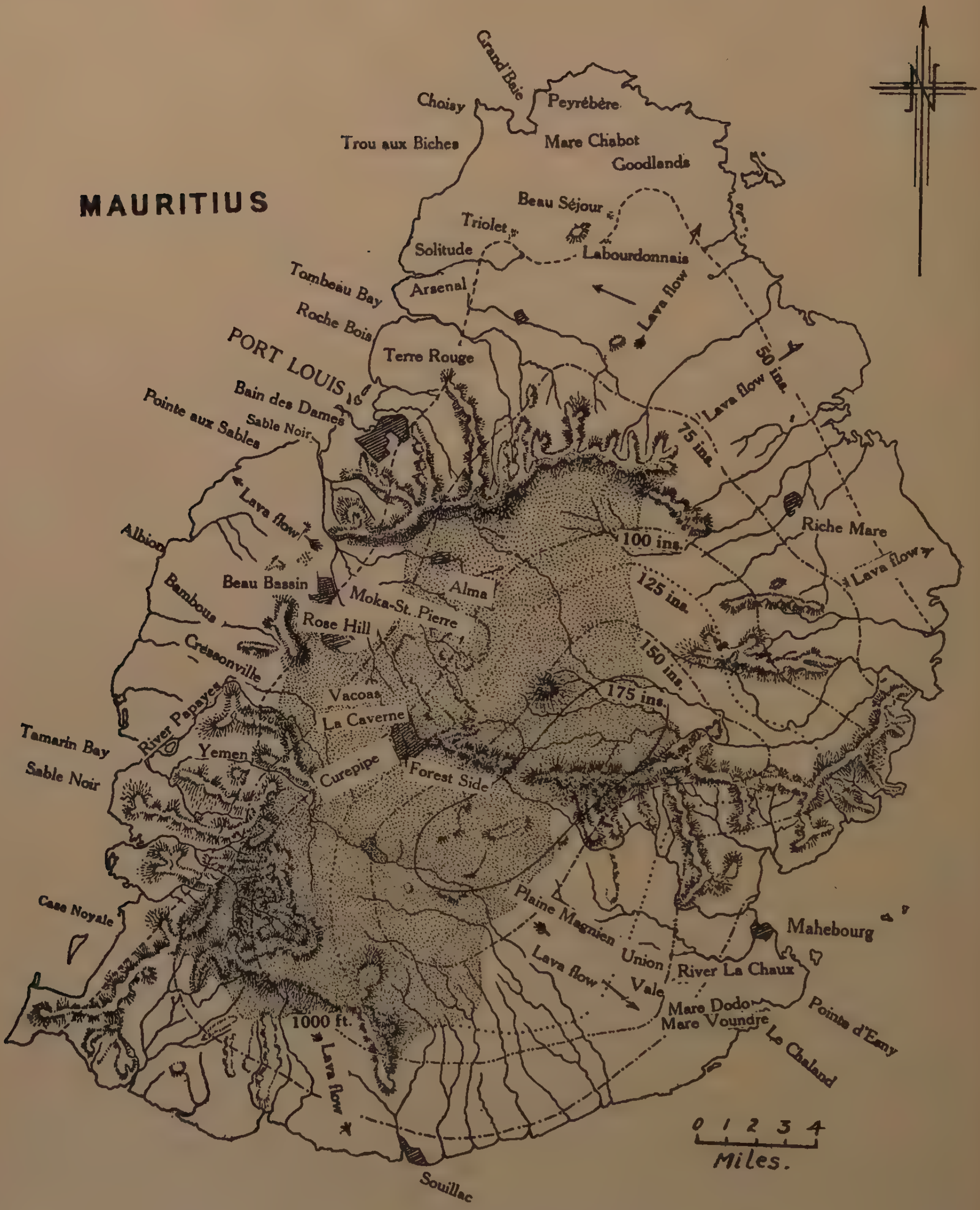
MAURITIUS
N
Grand Baie
Choisy
Peyrébère
Trou aux Biches
Mare Chabot
Goodlands
Beau Séjour
Triolet
Solitude
Labourdonnais
Tombeau Bay
Arsenal
Lava flow
Roche Bois
Terre Rouge
PORT LOUIS
50 ins.
Bain des Dames
Lava flow
Sable Noir
Pointe aux Sables
75 ins.
Lava flow
Albion
100 ins.
Riche Mare
Beau Bassin
Alma
Lava flow
Bambous
Moka-St. Pierre
125 ins.
Rose Hill
Cressonville
150 ins.
Vacoas
175 ins.
La Caverne
River Papayes
Tamarin Bay
Yemen
Curepipe
Forest Side
Sable Noir
Case Noyale
Plaine Magnien
Mahebourg
Union
River La Chaux
Lava flow
Vale
Mare Dodo
Mare Voundre
Pointe d'Esny
Le Chaland
1000 ft.
Lava flow
0 1 2 3 4
Miles.
Souillac

Fig. 1.

Introduction.

The investigations forming the subject of this paper were undertaken in order to elucidate certain specific questions arising out of large scale malaria control operations. In June 1942, a combined civil and military Malaria Control Unit was formed, under the direction of Dr. W. F. Jepson, Government Entomologist, to reduce the high incidence of malaria amongst Service personnel stationed in the hyperendemic coastal areas of Mauritius. The unit consisted of four sections each under the direction of a technical officer—(1) Medical survey (Dr. G. Sippe), (2) Water survey and routine Anopheline control (Mr. S. Gebert), (3) Entomological research and Anopheline density survey (Mr. A. Moutia), and (4) Bush-clearing, major anti-malarial works and bonification (Mr. E. Lesur in collaboration with the Admiralty Civil Engineer's Department).

Special stress has been laid on the necessity for close entomological control on the planning of all anti-malarial works, and the research programme has been closely coordinated with the needs of the other sections.

The Entomological Section has been the special charge of Mr. A. Moutia, Assistant Entomologist with H.Q. at the Laboratory of the Entomological Division, Department of Agriculture. A special staff consisting of two trained Technical Assistants and three mosquito searchers was engaged, the cost of the investigations being met by a special advance from the Mauritius Government and later by the Admiralty Civil Engineer's Department.

Short History of Anophelines in Mauritius.

The first evidence of the presence of malaria in Mauritius was the violent epidemic of 1865, in which over 10,000 persons perished, that started in the Black River District. It is a matter for speculation whether Anophelism without malaria existed prior to this outbreak, but in view of the large numbers of Indian immigrants in the two previous decades it is reasonable to assume that the two notorious African vectors *Anopheles gambiae*, Giles, and *A. funestus*, Giles, were accidentally introduced subsequently to the immigration, probably from the nearby Island of Madagascar. The discovery by Ross in 1898 of the transmission of malaria by *Anopheles*, gave added impetus to the study of the CULICIDAE. The names of d'Emmerez de Charmoy and Daruty de Grandpré will always be associated with these studies (Culicidae of Mauritius 1900).

The visit of Sir Ronald Ross and Major Fowler, R.A.M.C., in 1908 constituted the first attempt at a comprehensive study of malaria as an island-wide problem. *A. gambiae* was the only vector of importance then present. Unfortunately the recommendations of these workers were only carried out in part with the result that Sir Andrew Balfour in 1920 found the state of affairs such as to warrant a further intensive study. Mr. Malcolm MacGregor, an entomologist, visited Mauritius in 1922 and his report (1924) together with his book "Mosquito Surveys" (1927) are the basis on which all later entomological work has and should be founded. In particular his discovery that *A. funestus* was firmly established in Mauritius as a dangerous factor in malaria transmission completely changed the picture as regards methods of control. Since then, many interesting notes on Anopheline habits have been published by Mr. S. Gebert of the Medical and Health Department (1936, 1937, 1939), who has directed the field anti-Anopheline work of the present Malaria Control Unit, and who has unhesitatingly placed his long experience at the disposal of the present investigation.

Species of Mauritian Anophelines and their present Status.

A series of the four Mauritian species, including specimens of *A. gambiae* from saline coastal swamps, was submitted to Mrs. E. C. C. Van Someren of the Medical Research Laboratory, Nairobi, for examination. All the specimens proved to be typical forms and there was no evidence of the presence of *A. gambiae* var. *melas*. The following key modified from MacGregor (1922) and Evans (1938) is designed to serve the field worker for rapid identification of adults and larvae :—

Key to the Females of Mauritian Anophelines.

1. Large black or blackish brown species ; costa dark with two pale areas near the apex 4
2. Large (8 mm.) to medium sized (5–6 mm.) species ; costa with distinct black and white interruptions..5
3. Smaller (3–6 mm.) species with body colour distinctly brown ; costa with distinct dark and pale interruptions..6
4. Legs black with yellowish bands at articulations. Tarsi : 3rd segment half white half black. 4th and 5th segments entirely white. Palpi black with 4 pale bands very shaggily clothed with scales....................*A. coustani*, Giles (*mauritianus* Grandpré & Charmoy)
5. Palpi black, speckled with 3 pale bands. Legs speckled with I no pale speckling apically ; II apical half white ; III–V entirely white...*A. maculipalpis*, Giles
6. Medium to small brown species : average body length 5 mm. Distinct pale bands at tarsal joints on forelegs and nearly always on the other legs. Pale areas along wing fringes extend to termination of anal (6th longitudinal) vein......... *A. gambiae*, Giles (*costalis*, Loew)
7. Small to very small brown species : average body length 3–5 mm. Legs including fore tarsi without distinct pale bands. Pale areas along wing fringes but no pale area at termination of anal (6th longitudinal) vein......*A. funestus*, Giles

Key to Mature Larvae of Mauritian Anophelines.

1. Large species. Exterior clypeal hairs heavily branched and tufted ; thoracic hairs and anal gill hair tufts long and prominent. Palmate hairs large and distinct. Comb large composed of long and short teeth alternating (fig. 2)...... *A. coustani*, Giles (*A. mauritianus*, Grandpré & Charmoy)
2. Medium or small species. Exterior clypeal hairs unbranched and simple.........3
3. True palmate hairs present 5
4. Palmate hairs reduced to a brush or tassel form...6
5. Colour variable usually brown, sometimes greenish, with a distinct melanic variety which is pigmented. Head and thorax well separated. True palmate hairs present but inconspicuous. Abdominal tergites slightly chitinised. Combs small with fairly regular short teeth (fig. 2)............*A. gambiae*, Giles (*A. costalis*, Loew)
6. Small larvae (½ in.). Colour black. Head and thorax together appear like a triangular shield, thoracic margins appear transparent. True palmate hairs present and very conspicuous. Abdominal tergites heavily chitinised. Comb with long irregular teeth (fig. 2)..................................*A. funestus*, Giles
7. Brown larvae, resembling *A. gambiae* but palmate hairs reduced to tassel form. Comb large and irregular...*A. maculipalpis*, Giles

Field Recognition Characters.

For practical purposes in Mauritius, field recognition of the eggs and pupae of Anophelines or of all stages of *A. maculipalpis* should not be attempted except by an experienced worker. The comparative rarity of *A. maculipalpis* and the superficial similarity of the larva to that of *A. gambiae* render such attempts superfluous in day to day routine control. It is sufficient to say that larvae of *A. maculipalpis* are confined to pools resulting from natural seepages and should a mass of brown larvae be found in such a breeding place, they should be submitted to laboratory examination.

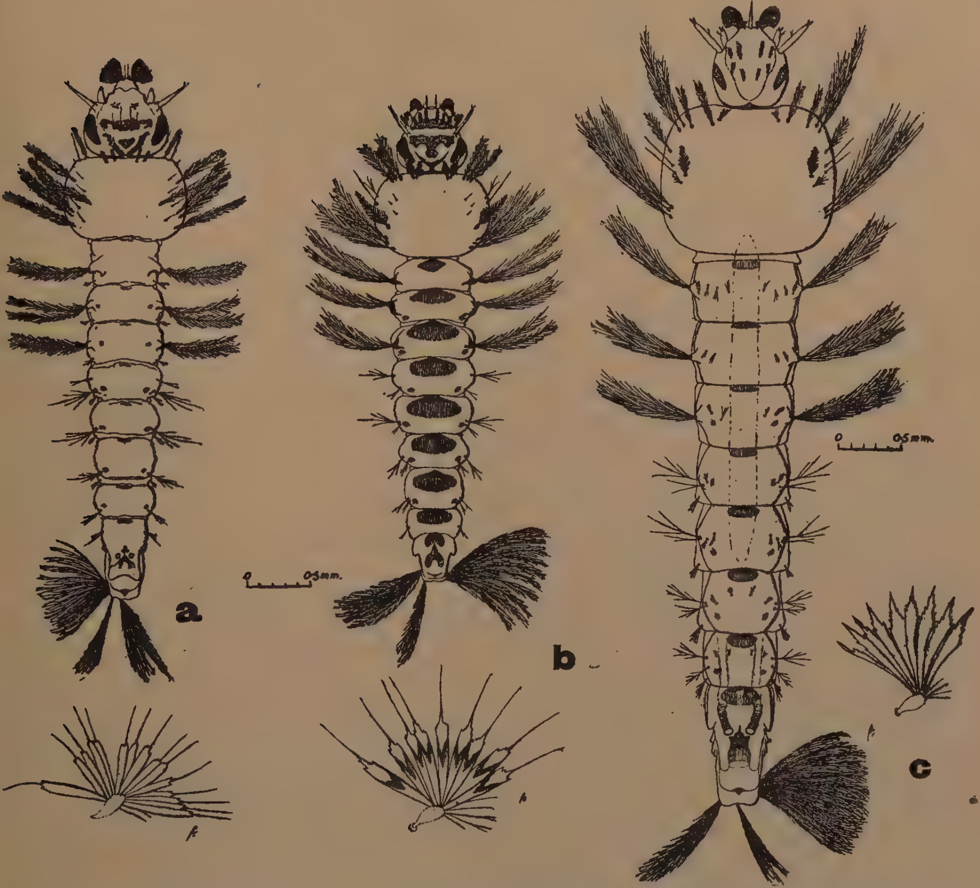

Fig. 2.—Mature larvae of: (*a*) *A. gambiae*, Giles; (*b*) *A. funestus*, Giles; (*c*) *A. coustani*, Giles.

The field characteristics of the three chief Anopheline species are as follows :—

A. coustani, Giles (*mauritianus*, Grandpré & Charmoy)

Breeding abundantly in the clean, preferably slowly, moving fresh water of grassy pools that are replenished either by rain or by the flow of a stream ; this species may also be found in natural waters in all parts of Mauritius from the coast to 2,300 feet. Some degree of pollution by rotting vegetation is tolerated. The young larvae appear linear, almost hair-like, but the mature larvae are robust and conspicuously larger than the other Mauritian species. The body and tail hairs are very conspicuous and the species is difficult to confuse, except in the case of small specimens, with *A gambiae*. On the lowlands where *A. coustani* is found, the presence of the shy species *A. funestus* may be suspected for the two have similar breeding places. The adult *A. coustani* is a large black insect with thick shaggy palps ; the female often bites during the day and is common in the non-malarious regions above 1,500 feet, but seldom enters houses and is seldom captured in routine spraying operations.

A. gambiae, Giles (*costalis*, Loew).

Breeding in all open sunny waters, this species has a considerable tolerance for pollution and salinity and is therefore to be found in domestic wastes and in crab holes and pools near the sea side, in addition to a host of natural breeding places such as marshes, rock pools and casual rainwater pools. Young larvae are black and frequently have a distinct whitish collar up to the third stage and are found in great density on the surface of sunlit pools. Mature larvae are brown to greenish with varying percentages of darker " melanics ", and smaller than *A. coustani*, with body hairs inconspicuous. They move rapidly with a large number of darting movements (*cf. funestus* below). The adults are brown medium-sized insects with distinct white markings on the legs, which serve to distinguish small specimens from *A. funestus*, and they are typically anthropophilous and nocturnal. *A. gambiae* may, though rarely, be abundant at places over 1,000 feet above sea level.

A. funestus, Giles.

Breeding for the most part in natural waters covered by clean grass or vegetation ; the larvae are quite intolerant to salinity. Typical breeding places are Songe marshes (*Alocasia macrorhiza*, Scholl), grassy pools on flooded river banks, grassy irrigation channels with slow moving water and frequently in shady grass-free pools containing heaps of old rotted leaves and branches. Only one instance (Tombeau Bay) of breeding in an artificial basin 20 ft. × 20 ft. has been recorded. The larvae are somewhat difficult to find except in very heavy infestations, since they dive to a depth of several inches at any unaccustomed vibration. Young larvae are minute, quite black and difficult to diagnose in the field except from their situation, but mature larvae are quite black and appear dry on the dorsal surface owing to the effect of the large palmate hairs in bringing the body above the water surface. The shape of the thoracic shield, formed by the head and thorax together, is typically triangular with the thoracic edges somewhat translucent. Movements of *A. funestus* larvae are typically more abrupt and less continuous and extensive than those of the other species. The adult mosquito is usually identifiable by its small size (about $\frac{1}{8}$ in.) compared with that of *A. gambiae* which it greatly resembles morphologically. The legs of *A. funestus* are, however, quite devoid of white markings of any kind. Wing characters can only be seen under a powerful lens. Adults are commonly found in human habitations in the coastal areas at distances of $\frac{1}{2}$ to 1 mile from their nearest breeding places. They have been found up to 1,500 feet (Alma) in the present investigation.

Anopheline Surveys : Aims, Methods and Results.

The aims of the survey were threefold :—

1. To obtain information on the incidence of Anophelines in areas to be controlled so as to enable work to be planned and priorities allotted.

2. To assess the progress of control and to lay the foundations for permanent records.

3. To elucidate such facts on the relative incidence of Anophelines in houses and huts at different seasons and times as were necessary to correlate entomological work with the findings of the malaria survey amongst the civil population by the Medical Section under Dr. G. Sippe.

Method of Assessment of Anopheline Abundance.

The method of assessing abundance by establishing fixed catching stations where regular searches for mosquitos are made was rejected after considerable study. Nearly all small human habitations in Mauritius are of two patterns, the Indian wattle and daub type thatched hut consisting of 1 or 2 rooms of nearly uniform size (1,440 cu. ft.), or the creole or European board or petrol tin house with corrugated iron roof with rooms of size similar to the above. Both types are capable of being sealed reasonably well by means of cyclone shutters. Spray gangs armed with Flit guns and white sheets were trained to carry out a thorough spraying of the closed huts with standard PYEX (pyrethrum spray), and to count and identify the mosquitos collected on the sheets and on the smooth floors, by the light of electric torches. The time spent on each hut did not exceed 10 minutes and even allowing for an estimated loss of 25 per cent. by insects escaping, the survey of a relatively large number of huts at random gave a better result than the less extensive and no more accurate work of the mosquito searcher. Furthermore, the co-operation of the population was assured on account of the popularity of an operation that killed the numerous other denizens of the gloomy huts.

Morning and Evening Spraying.

At first only day spraying, between 7 a.m. and 3 p.m., was carried out owing to staff and transport difficulties but with the creation of local resident maintenance gangs, evening and night spraying became possible. Comparative sprayings were carried out in the same huts by day and by night after it had been established that no residual repellent effect was produced after 48 hours by the dosage of pyrethrum applied in the normal course of hut spraying operations.

The results of the two series of observations were as follows :—

Series 1.—Cassis, Port Louis. 79 rooms were sprayed between 0600 to 0830 and 1800 to 2030 hours from 16th to 30th March, 1944 (malaria season). There was an interval of 48 hours between sprayings. The morning and evening catches for *Culex* were 2,724 and 1,064 respectively and for *Anopheles gambiae* 76 and 17.

The spraying was repeated in 89 rooms in September 1944 (non-malaria season) when the morning and evening catches were for *Culex* 266 and 207 respectively and for *Anopheles* nil.

Series 2.—Mon Desert Estate, Moka (1,200 ft.). 2 rooms were sprayed between 15th and 27th February, 1943, from 0630 to 0830 hours and from 1830 to 2030 hours. The morning and evening catches of *A. funestus* for 6 days were 104 and 246 respectively and for *A. gambiae* for 12 days, 90 and 20.

In general, morning sprayings gave the greatest number of *A. gambiae* and Culicines ; whereas *A. funestus* was always more abundant in the early evening sprayings. It was constantly observed that collections of live mosquitos for dissection made after 9 a.m. were practically all *A. funestus*, and that if *gambiae* were required, searches had to be made in dark huts in places where the density of infestation was very high. In practice, special stress has been laid on training troops and other personnel in the careful use of anti-mosquito clothing in the early morning as well as at sundown.

Distribution and Seasonal Occurence of Anophelines in Mauritius.

Limits of Area surveyed.

The surveys did not cover the whole of Mauritius, the Districts of Riviere du Rempart and Flacq in the east and of Savanne in the south being excluded from the normal field of action of the Malaria Control Unit. However, the area covered (see fig. 1) includes practically every type of mosquito habitat likely to be encountered.

Importance of A. funestus.

It is not far wrong to state that wherever splenic indices of the order of 75 to 100 per cent. occur, *A. funestus* will be found in considerable abundance (10 to 350 adults per room). Such places will always be found on inspection to be surrounded by natural breeding places such as uncontrolled marshes, untrained rivers or neglected irrigation systems. Wherever urbanisation has proceeded, or partial anti-malaria work has been carried out (*e.g.* Grand'Baie), *funestus* gives place to *gambiae* and the incidence becomes fairly strictly confined to the 4 or 5 months of the hot wet season, with a corresponding lessening of the severity of incidence. The following table illustrates this important point :—

TABLE I.

Place	Splenic Index	Chief Vector Species	Max. *Anopheles* catch per room 1942–43	Transmission period	Type of Breeding Place found
Albion, Black River ...	100	*funestus*	335	10/12	Freshwater coastal reed swamps.
Montee des Hussards, Grand River N.W.	95	*funestus* and *gambiae*	143	8/12	"Songes" pools + Dayot canal, saline marshes.
Tamarin, Black River ...	94	*funestus*	151	8/12	Weedy banks of sluggish river estuary.
Mon Desert, Mahebourg ...	89	*funestus*	97	8/12	Factory marsh + River les Marres.
Pailles	87	*funestus*	57	8/12	St. Louis river.
Cassis, Port Louis, 1942 ...	67	*funestus* and *gambiae*	53	7/12	Dayot irrigation branches. Water pools. Salt pan.
Grand'Baie	49	*gambiae* and *funestus*	79	4/12	Large open marshes.
Cassis, Port Louis, 1943 ...	38	*gambiae*	6	3/12	Casual water in summer.
Cannoniers Point	35	*gambiae*	24	3/12	Large partially filled marshes without reeds
Beau Séjour	33	*gambiae*	10	2/12	Man-made nuisances near village.

" Songe " is a swamp aroid.

Anopheline Density and Malaria.

On the above basis of Anopheline density, it has been observed in practice that habitations or barracks that yield 1 to 5 Anophelines per hut are regarded as fairly highly malarious. In December 1943, an interesting case occurred of a single unoiled pool causing a flush of *A. gambiae* in a cantonment of over 100 huts each of 3 rooms. The frequency distribution of male and female mosquitos in 35 huts selected at random showed an average of 2·63 males (±·433) to 4·86 females (±·375).

This flush of Anophelines produced malaria cases amounting to 10 per cent. of the number of occupants within a period of 2 weeks.

Vertical Distribution.

The findings of this investigation on vertical distribution agree closely with those of MacGregor except that *A. funestus* has been found in abundance up to 900 feet and in one circumscribed epidemic area at 1,500 feet (Alma). In typical *funestus* breeding places, abounding in upland marshes, ponds and ditches from 900 to 2,300 feet, nothing but *A. coustani* has yet been found. Possible reasons for this will be discussed in a later section. In the case of *A. gambiae,* a rapid decrease in density occurs as the central plateau is reached and above the 1,000 ft. contour (upper parts of the centres of Beau-Bassin, Rose-Belle and Quatre Bornes) only occasional specimens can be found, with rare local concentrations in particularly favourable spots. But in spite of widespread and frequent searches of waters outside the townships where bonification is well advanced, *A. gambiae* has not been encountered during the present work, and it must therefore be concluded that some natural controlling factor, such as low temperature, lack of suitable algal or bacterial food, or flushing of streams and pools by heavy rains, must be operative in these regions.

" MacGregor Line."

For most practical purposes, therefore, the principle of the " MacGregor line", adjusted to between 900 and 1,000 feet above sea level for *funestus,* may be retained to give a broad picture of Anophelism and malaria in Mauritius. It is impossible to say whether there has been a progressive advance of Anophelines into the plateau areas, since the time of MacGregor, but the spleen rates indicate little, if any, general change in malaria incidence since Ross' survey in 1907, except in local areas such as Clairfonds (Phoenix) where the spleen rate of over 80 per cent. was speedily reduced to about 3 to 5 per cent. by marsh drainage in 1908.

Effect of Environmental Factors upon Anophelines in Mauritius.

Since malaria incidence in any locality is nearly always dependent upon environmental factors favourable to the mass increase of the vector, some attempt has been made to evaluate these factors and to assess the part they play in producing the irregular distribution of the disease observed in the Colony.

It must be noted that the statistics quoted are derived mainly from field observations. The difficulty of reproducing natural conditions in the laboratory, particularly those of nutrition and probably oxygenation of breeding water, introduces many unknown variables and suggests that the statistical findings should be regarded merely as an aid to the observational or subjective findings.

The factors considered below are : (1) Temperature. (2) Rainfall and humidity. (3) Soils. (4) pH and salinity etc. of breeding water. (5) Melanism in *A. gambiae* larvae and salinity. (6) Light. (7) Wind.

1. *Temperature.*

Within the limits imposed by the availability of water, temperature is the factor of limiting importance.

(a) *Temperatures normally occurring in Mauritius.*

Table II shows the mean temperature data for typical coastal plain (Pamplemousses Observatory) and upland (Curepipe) stations (*i.e.* malarious and non-malarious).

The warmest mid-day temperatures rarely exceed 30°C. in January and February. Measurements of the corresponding temperatures in shallow sunny pools have varied from 30° to 39°C. with a mean of about 32°C. In the uplands, however, water temperatures above 25°C. are rare and the usual mid-day reading in a typical *gambiae* pool is from 23° to 24°—more than 10°C. below the optimum breeding temperature.

TABLE II.

	Pamplemousses Observatory (150 ft.)			Curepipe (1,800 ft.)		
	Mean monthly Max. Temp. °C.	Mean monthly Min. Temp. °C.	Mean monthly Temp. °C.	Absolute monthly Max. Temp. °C.	Absolute monthly Min. Temp. °C.	Mean monthly Temp. °C.
January	29·8	22·6	25·89	29·2	17·4	23·0
February	29·5	22·8	25·81	29·8	15·7	23·1
March	28·9	22·3	25·22	28·2	16·8	22·4
April	27·6	21·1	24·11	27·3	15·5	21·2
May	25·9	19·1	22·20	25·9	11·4	19·7
June	24·4	17·0	20·48	24·2	10·0	17·6
July	23·7	15·5	19·82	22·8	10·9	17·5
August	23·9	16·5	19·96	23·2	9·6	15·5
September	25·1	17·0	20·78	23·5	10·1	18·1
October	26·8	18·0	22·11	25·1	12·4	18·8
November	28·4	19·6	23·72	26·1	12·8	19·9
December	29·7	21·6	25·29	28·7	14·4	22·1

TABLE III.

Life-cycle of *Anopheles gambiae*—1944.

Place	Bain des Dames	Fort William	Bain des Dames	Canote	Bain des Dames
District	Port Louis	Port Louis	Port Louis	Black River	Port Louis
Time of year ...	March 1944	March 1944	May 1944	July 1944	Sept. 1944
Type of breeding place	Natural pool	Pool dug near sea shore	Pool dug in secondary drainage channel	Edge of small stream	Pool dug near salt pans
Conditions of breeding	Breeding done inside tin fixed in soil	No tin used	Breeding done inside tin fixed in soil	Breeding done inside tin fixed in soil	No tin used
Specimens used ...	1st stage larvae	1st stage larvae	eggs	1st stage larvae	1st stage larvae
Salinity (NaCl) ...	0·6 grs. per litre	17·5 grs. per litre	9·7 grs. per litre	0·2 grs. per litre	15 grs. per litre
pH	8·0	8·4	8·2	7·8	8·3
Period from Egg to larvae...	1 day	1 day	1 day	1 day	1 day
Larvae to pupae	6 days	6 days	11 days	19 days	10 days
Pupae to adults	1 day	1 day	1 day	1 day	1 day
TOTAL ...	8 days	8 days	13 days	21 days	12 days
Mean Water Temp.	32·8°C.	28·9°C.	26·0°C.	23·0°C.	25·3°C.
Max. Water Temp.	36·0°C.	31·8°C.	28·0°C.	—	28·2°C.
Min. Water Temp.	29·0°C.	27·0°C.	24·0°C.	—	21·5°C.

TABLE III—*continued.*

Life-cycle of *Anopheles gambiae*—1945.

Place	Cassis	Cassis	Fort William	Fort William	Fort William	Malaria Section
District	Port Louis	Port Louis	Port Louis	Port Louis	Port Louis	Vacoas
Time of year ...	March 1945	March 1945	March 1945	March 1945	March 1945	March 1945
Type of breeding place	Pool made in small seepage drain	As last	Concrete trough divided in sections by mud walls	As last	As last	Dishes 12×9 ins.
Conditions of breeding	Breeding done inside tin fixed in soil	As last	Shallow muddy water algae added	As last	As last with cage over water	Intermittent sunlight
Specimens used...	1st stage larvae	Eggs	1st stage larvae	Eggs	Gravid ♀♀	Eggs
Salinity ...	1·0 grm. per litre	1·0 grm. per litre	Tap water	Tap Water	Tap water	Tap water
pH	Not taken	Not taken	Not taken	Not taken	Not taken	Not taken
Period from Egg to larvae...	1 day	1 day	1 day	1 day	1 day	1 day
Larvae to pupae	7 days	11 days	8 days	6 days	7 days	13 days
Pupae to adults	2 days	2 days	1 day	2 days	2 days	2 days
TOTAL ...	10 days	14 days	10 days	9 days	10 days	16 days
Mean Water Temp.	29·6°C.	28·1°C.	31·4°C.	30·8°C.	31·7°C.	25·5°C.
Max. Water Temp.	36·0°C. (p.m.)	36·5°C. (p.m.)	35·0°C.	37·0°C.	38·0°C.	27·0°C.
Min. Water Temp.	24·0°C. (a.m.)	25·0°C. (a.m.)	24·0°C.	26·5°C.	28·0°C.	23·0°C.

Furthermore it is significant that the number of months in which the mean monthly temperature is under 20°C. is :

2 for Pamplemousses
5 for Vacoas
7 for Curepipe

At Curepipe (1,500 ft.) the absolute minimum temperature is below 16°C. for 10 months of the year, and in August minima of 9° to 10°C. are commonly recorded. The effect of these temperatures, which show a regular gradation at stations of intermediate heights, on the developmental period of *A. gambiae* will now be considered.

(b) *Effect of Temperature on the Development of* Anopheles gambiae *and* funestus.

(i) *A. gambiae.*

The life-cycle of *A. gambiae* has been worked out in the field at several different seasons, using natural breeding places as a check on careful laboratory rearings, thus avoiding the largest source of error since laboratory rearings tend to give values for the life-cycle period well in excess of those observed in nature. Pools, where dug, were placed adjacent to existing free breeding pools. Table III gives the details of these experiments. Temperatures were recorded in most cases thrice daily but these

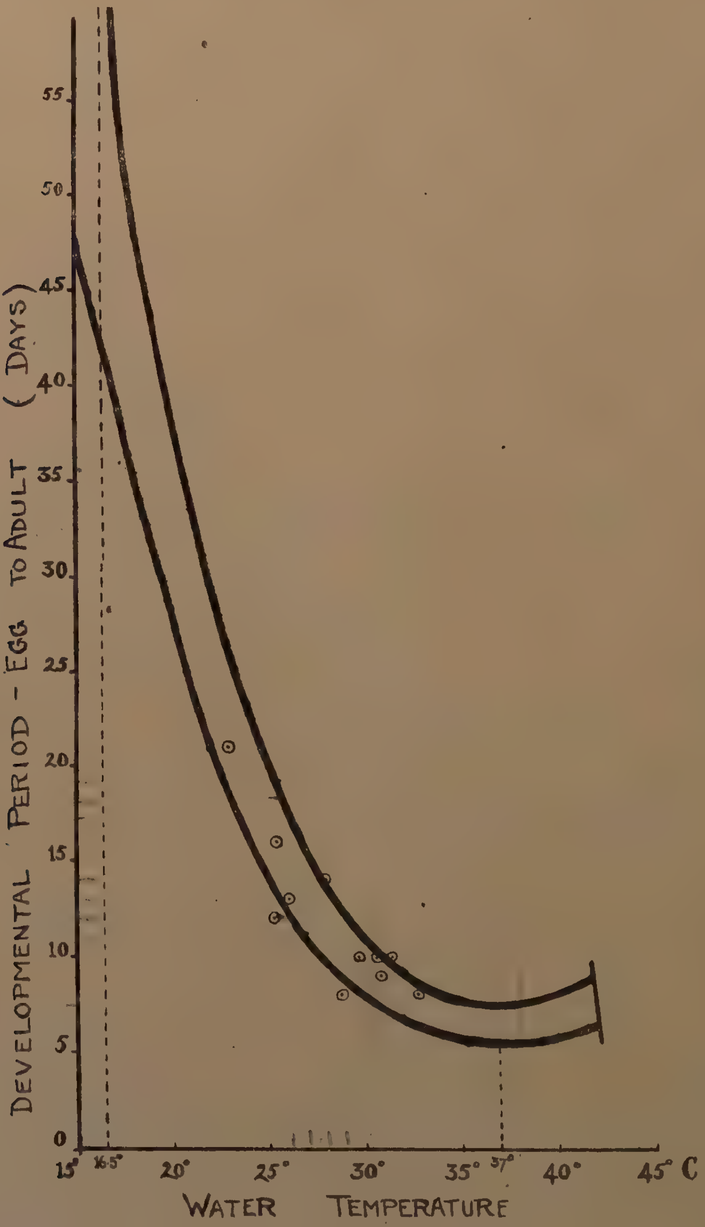

Fig. 3.

may tend to be on the high side owing to absence of night readings. However, when plotted, the points lie approximately on a curve usual to this type of work (fig. 3) a hyperbolic or more probably an exponential function. In order to complete a theoretical curve of temperature development the minimum developmental period for *A. gambiae* must be known.

Some idea of the upper lethal limit for larvae was obtained by a series of laboratory observations (4 replications each of 20 larvae).

TABLE IV.

Species	Initial Temperature	Temperature of water during Treatment	Duration of Treatment	Percentage larvae dead
A. gambiae, Loew	25°C.	40°C.	4 hours	nil
		41	1 ,,	nil
		41	4 ,,	nil
		42	¼ ,,	30
		42	½ ,,	45
		42	1 ,,	75
		42	2 ,,	100
		43	¼ ,,	100
A. funestus, Giles	27°C.	38°C.	4 hours	nil
		39	1/10 ,,	nil
		39	¼ ,,	10
		39	1 ,,	20
		40	¼ ,,	10
		40	½ ,,	30
		40	¾ ,,	55
		40	1 ,,	80
		40	1¼ ,,	100

The limit for *A. gambiae* is thus about 41°C., or some 2°C. above that for *A. funestus*. It can be shown that the shortest (though not necessarily optimal) developmental period in many insects, if not all poikilothermal organisms, occurs at temperatures within 3 or 4 degrees of the thermal death point (Janisch 1931, Maercks 1933). With a field knowledge of the tolerance by larvae of temperatures up to 39° in natural pools a reasonable estimate of the minimum of the curve is 37°C. A theoretical symmetrical catenary, or more properly two catenaries, can now be drawn with the following formulae :—

$$y=2{\cdot}75\ (1{\cdot}144^{x}+1{\cdot}144^{-x}).$$

$$y=3{\cdot}75\ (1{\cdot}148^{x}+1{\cdot}148^{-x}).$$

The area so enclosed contains nearly all the observed values and is offered as a suggestion for the behaviour of the species with respect to water temperature, although it is probable that an asymmetrical catenary would give an even better picture. This cannot, however, be produced until further projected work has been done at superoptimal temperatures in natural breeding places. In Mauritius some degree of artificial heat or heat insulation would be essential to maintain such temperatures even in the warmest pools.

It can be seen at once that whilst temperature variations between 30° to 38° are of minor importance in modifying the optimal conditions of development this is far from being the case in the range 18° to 26°, where a decrease of 1°C. in temperature may add a week or more to the developmental period. Furthermore, the curve demonstrates the observable fact that at some temperature not lower than 18° or 19°, the life-cycle of *A. gambiae* may be prolonged to well over one month. Since it is well known that the symmetrical catenary curve errs towards the low side at extremes of low temperature, it is apparent that at the temperatures occurring in Mauritius in winter on the coast and over a considerable period of the year upcountry, *A. gambiae* will rarely be able to complete its life-cycle. In the laboratory, a series of tests with *A. gambiae* showed that all activity of the larvae ceased at 16·5°C. and although the theoretical curve shows no asymptote, the period of from 45 to 60 days is, for practical purposes of malaria transmission, a near equivalent to infinity. Only in very special situations such as permanent marshes fed by a large river would the water and food supply conditions remain favourable for continuous periods of 1 or 2 months at a time. In the vast majority of available breeding places drying, pollution, flushing and predators would intervene as limiting factors.

The fact remains, however, that in the present hot season (March 1945) *A. gambiae* has been reared from eggs at a station 1,300 ft. above sea level at a mean natural water temperature of nearly 25° in 16 days. It is, therefore, possible that some trophic factor such as the precipitation of food proteins in natural waters by the presence of some metallic ion such as iron or aluminium must be added to that of temperature in order to provide an explanation for the observed absence of the species from such a wide upland area in the Colony.

(ii) *A. funestus.*

As regards *A. funestus*, the normal temperatures of the breeding places under the lightest shade of grass rarely exceeds 25°–26°C. and the developmental period is rarely under 21 days. In the laboratory, at the height of summer (Feb. 1944) at a mean temperature of 24°C. the total life-cycle varied from 20 to 31 days, with a mean of 24 days. Under optimal environmental conditions in nature this figure might be reduced by a few days, but the life-cycle remains at nearly three times that of *A. gambiae*. This may explain how the species can easily be completely controlled in any area by the abolition of the permanent type of breeding place. The complete control of *A. funestus* is of fundamental importance in the malaria problem. Further critical work is necessary regarding the common assumption that heavy shade will of itself banish *A. funestus*. In this laboratory it is believed that the temperature lowering effect of shade is more important than the lack of light, and further consideration is given to this important question later.

2. *Rainfall and Humidity.*

(a) *General considerations.*

Rainfall studies in Mauritius are complicated by the great variations that occur within a distance of as little as one mile and the fluctuations of the mean rainfall for any particular month over a period of years may exceed 50 per cent. of the mean.

There are two principal ways in which rainfall affects the abundance of Anophelines :—

(i) The annual wet season rains from December to April, both orographic and cyclonic, fill the marshes and hollows, causing seepages on sloping ground and a sustained increase in the level of rivers. As a consequence, the number of available breeding places is multiplied.

(ii) Periodical cyclonic depressions produce phenomenal rains amounting to as much as 30 inches in 48 hours. The resultant spates produce an effective flushing effect upon rivers that would normally be serious breeding places.

(b) *Distribution of Rainfall and Malaria.*

Isohyets are indicated on the Map (fig. 1). Broadly speaking rainfall in the warm lowlands ranges from 60 to 80 inches on the weather (S.E.) side of the Island, the whole of the West Coast and the Northern Plain varying from 30 to 50 inches. The higher rainfalls of 80 to 175 inches occur at levels that are free from vector species and only affect the lowlands because this vast volume of water is drained into or through them by underground rivers or by deep ravines. There is little significant correlation between rainfall distribution and the corresponding splenic indices in coastal areas where permanent swamps abound ; on the inland plains (more than

3 miles from the coast) low rainfall means less malaria, with a definite short season of occurrence. Table V illustrates this (complications due to irrigated lands have been eliminated) :—

TABLE V.

Malaria and Rainfall in Inland Plains below 500 feet.

Station	District	Height in Feet	Mean Annual Rain	Splenic Index 1943
Triolet	Pamplemousses (North) ...	100	40	17
Goodlands	Riv. du Rempart (North) ...	100	50	10
Labourdonnais Estate ...	Riv. du Rempart (North) ...	250	58	25
Queen Victoria Estate ...	Flacq (East)	400	90	?
L'Etoile Estate	Flacq (East)	450	120	87
Plaine Magnien	Grand Port (South)	250	75	60
Terracine Estate ...	Savanne (South)	50	70	94

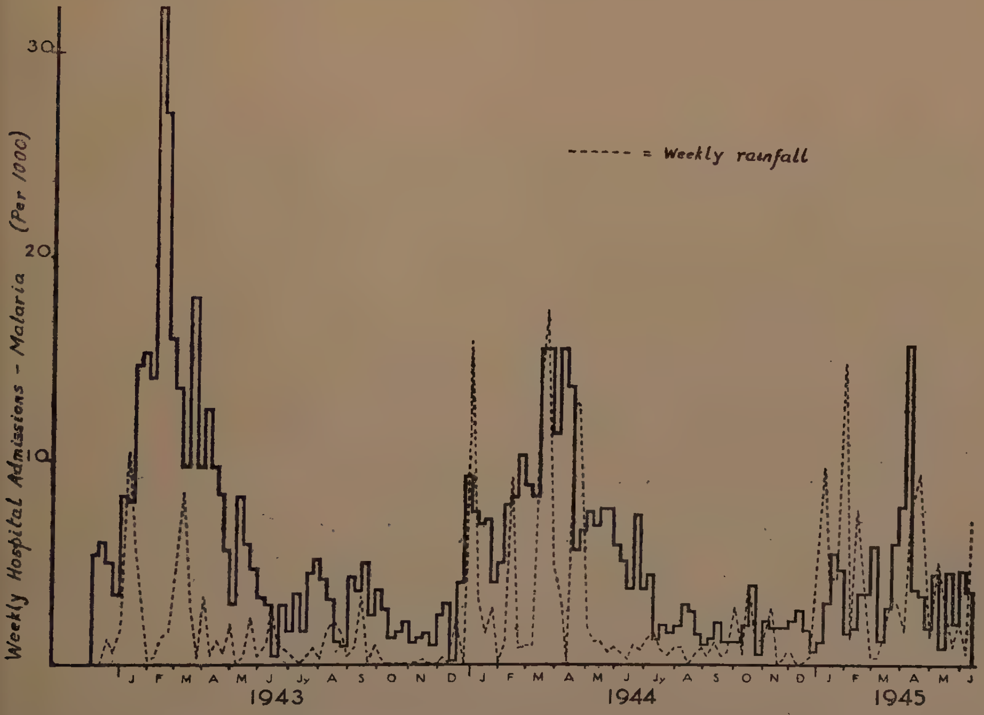

Fig. 4. Rainfall and Malaria Incidence amongst Service Personnel.

The proximity of the higher rainfall stations to catchment areas and their consequent intersection with ravines and other drainage channels undoubtedly plays a part in raising the level of malaria incidence. Although *Anopheles* may be very abundant for a few weeks following falls of rain exceeding 6 inches, over a period of 2 or 3 days, it is difficult during the months of May to November to find a specimen in the dry northern area. In these areas the spleen index is generally 25 to 40 per cent. and the mean hut infestation with *A. gambiae* is under 5. The corresponding figures for the higher rainfall areas are 60 to 100 per cent. spleens and 15 to 20 *A. gambiae* together with numbers of *A. funestus* running into hundreds.

There are well marked seasons of heavy and light precipitation. The rainfall by weeks for the past 3 years in an important controlled area (100 ft.) is shown in fig. 4. Daily precipitation is, of course, the criterion on which immediate oiling measures must be determined. Superimposed upon the rainfall diagram is the total weekly malaria incidence (per 1,000) amongst Service personnel. The general reduction in incidence largely consequent upon mosquito control measures, will be noted. The time lag between heavy rain and peaks of incidence is somewhat irregular. In 1943 at the beginning of the wet season, an exceptional peak of malaria incidence occurred in the week ending 13th February. The only rains of any significance at this time, fell in the first days of January, *i.e.* 5 to 6 weeks before. A similar interval was noted in February 1944, when a severe peak occurred 5 to 6 weeks after the heavy rains of 9th February. On the other hand, the early upswing of incidence in January 1944 appears to have no causative rains and is presumably due to an early onset of warm weather. The first heavy rains in January 1944 were actually followed by a considerable temporary decrease in malaria 5 weeks later, and this was repeated in 1945 after the cyclonic rains of 15th to 18th January. Since the chief vector at this season and in control areas is *A. gambiae*, there is evidently a conflict between the creation of widespread breeding places by these heavy rains and the flushing of these same places. In winter there appears to be some relation between showers occurring in the coastal areas and resulting peaks of malaria, but, on the whole, *A. gambiae* becomes confined to areas where permanent swamps or sluggish streams exist. These areas are well

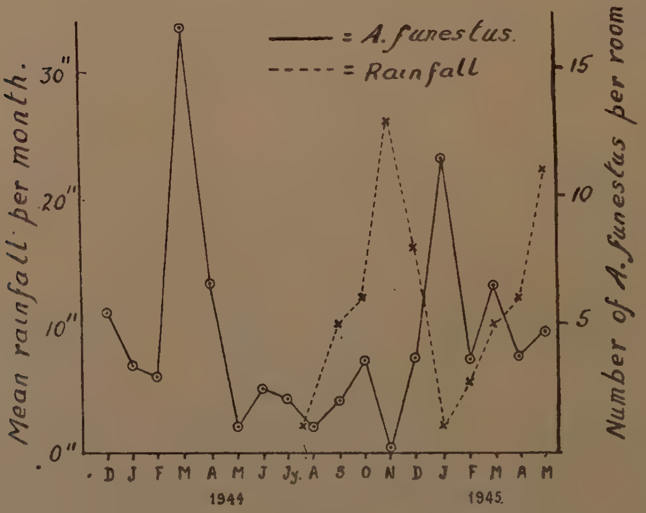

Fig. 5.

known to the local population and are shunned as holiday resorts. Accurate work on the correlation of Anopheline populations with variations in the quantity and quality of rainfall can alone enable malaria incidence to be predicted with any degree of certainty. The latest work of Ribbands (1944) has demonstrated the complexity of the rhythm of Anopheline populations occurring in human habitations, and it is obvious that only a series of observations made under standard conditions for a number of years will enable any great advance to be made along these lines. The effect upon catches of *A. gambiae* is also well marked, the upswing occurring 10 to 15 days after the rains, high catches persisting until 2 or 3 weeks after the drying of casual water in the area. Catches of *A. funestus* are not so dependent upon the rains, in fact the calmer period for 3 or 4 months after heavy rain appears to be the season of maximum abundance (fig. 5).

There is one special case in which a prolonged dry spell during the hot rainy season allows the rivers to become dangerously low and sluggish. The resultant heavy increase in Anophelines, particularly of *A. gambiae*, has, in the past, been the cause of local epidemics. In 1934, a very dry year, an outbreak at Reduit (900 ft.) was caused in this way. It is not improbable that the dry conditions in 1941–42 were responsible for an exceptional outbreak at Alma, Moka (1,500 ft.), in that they failed to flush the numerous ponds and marshes, a process which happens frequently in a normal year with a rainfall of 125 ins.

(c) *The flushing Effect of heavy Rains.*

The frequent spates that occur in the rapidly flowing streams of Mauritius, particularly in the uplands, are a factor of great importance in the natural control of Anophelines. It has, in fact, proved impossible, either in 1944 or 1945, to rear a generation of *A. gambiae* in a typical upland stream. Falls of 1 in. are sufficient to flush the average upland stream, but in order that the effect may become marked in the larger sluggish rivers as they near their mouths, rainfall of upwards of 4 to 6 ins. in the 24-hour period are necessary. Such conditions occur 3 or 4 times annually, at intervals of from 2 to 4 weeks of the critical Anopheline breeding season. An excellent example was observed in March 1945, when it was discovered that the growth of *Spirogyra* for ¼ mile at the mouth of the St. Louis river south of Port Louis was supporting a population of *A. gambiae* sampled at 50 larvae per dip. Arrangements were made to dust the area, but on the night of 8th–9th March over 4 ins. of rain fell in a few hours. The resulting spate caused a rise of 8 ft. near the mouth and on the following day not a single larva was to be found. The adult population of *A. gambiae* that had risen sharply from 10 to 450 (per 10 huts) fell within two weeks to under five. In this manner sharp local outbreaks of malaria may be produced in almost any region of Mauritius and may be terminated just as suddenly by natural flushing.

(d) *The Effect of Humidity on Anophelines.*

Atmospheric humidity during the hot wet season is constantly above 85 per cent. Although no exact studies have been made, experience in handling adult mosquitos shows that an atmospheric humidity of over 90 per cent., providing optimum humidity conditions for existence and long flight, will only occasionally be reached. Malaria transmission in the wetter lowlands is thus favoured, and will only rarely be optimal in the drier northern areas during exceptionally wet periods, as in fact happened in March and April 1944 at Beau Séjour and other northern Stations.

3. *Geology and Soils as Environmental Factors in Anopheline Occurrence.*

Mauritius is wholly volcanic and consists of superimposed grey-blue basaltic lavas of various ages, the typical sharp hill features being the remnants of harder darker lavas, some of which approximate to oceanites with much porphyritic angite and olivine. Areas of newer lava flows (see fig. 1) are characterised by lack of large rivers and in these areas, underground springs and seepages are particularly abundant. It is, however, of fundamental importance in Anopheline ecology that the soils of Mauritius are generally highly porous with degrees of tropical laterisation dependent roughly on rainfall (Craig 1934). Except, therefore, near sea level or in the uplands where areas of solid lava pan exist, casual water persists only for short periods except where seepages occur, and problems of elaborate and extensive surface drainage are uncommon. There are, however, a number of well defined areas of grey alluvial soils derived from highly magnesium oceanites, that are practically impervious, and that call for special drainage measures. The Bell Village area of Port Louis is the best example of this soil type.

The only other soil type which requires mention, and further special study, is that derived under high rainfall conditions from highly ferrugineous lava (coarse grained vesicular olivine basalt) and sometimes from beds of volcanic ashes and is typical of the uplands above 1,200 ft. Weathering of this rock in stream beds produces much iron precipitate of a colloidal nature. It has been observed that the growth of *Spirogyra* is in some way inhibited in such waters, and *A. gambiae* has never been found in them during the present investigation, though Mr. Gebert has reported it from time to time in past years. It remains to be determined whether larval food of this species is precipitated or otherwise destroyed by physico-chemical action.

4. *pH and Salinity of Breeding Waters.*

Special attention has been paid to this question in view of what has been said above concerning possible chemical action, and in view of the well known tolerance of *A. gambiae* for salinity.

The chief points of practical importance were to ascertain :—

(1) The limits of pH of naturally occurring waters in relation to Anopheline breeding.

(2) To what extent mangrove and other coastal swamps are dangerous as sources of the saline tolerant *A. gambiae.*

One hundred Anopheline breeding places were examined at random for species, pH and NaCl content. The results (Table VI), derived mainly from coastal situations, indicate that natural waters in the malarious lowlands are mainly on the alkaline side with a range of pH 7·0 to pH 8·6, the latter figure being due to admixture with sea water. It is noteworthy that the upland waters tend on the whole to be neutral or even slightly acid and, whilst this in no way affects *A. coustani*, it may be closely correlated with the general absence of the two vector species *A. gambiae* and *A. funestus*. There are, however, the temperature and rainfall factors to complicate the problem in upland areas.

TABLE VI.

Range of pH+Salinity in Natural Breeding Places.

Locality	District	Date	7·0	7·2	7·3	7·4	7·5	7·6	7·7	7·8	7·9	8·0	8·1	8·2	8·3	8·4	8·5	8·6	Chlorides grs. NaCl per litre	Species of *Anopheles**
Roche Bois	Port Louis ...	Sept. 42														+			33·9	nil (sea water)
Roche Bois	Port Louis ...	Dec. 42														+			30·5	nil
Roche Bois	Port Louis ...	Dec. 42										+							27·5	nil
Pte. d'Esny	Grand Port ...	Jan. 44													+				27·0	nil
Bain des Dames	Port Louis ...	Sept. 42																+	26·0	nil (sea water)
Tamarin	Black River ...	Nov. 43											+						25·7	G
Bain des Dames	Port Louis ...	Dec. 42												+					25·5	nil
Pte. d'Esny	Grand Port ...	Jan. 44													+				25·3	nil
Roche Bois	Port Louis ...	Sept. 42														+			23·5	G
Sable Noir	Port Louis ...	July 43													+				19·8	G
Pte. aux Sables	Black River ...	Feb. 44														+			19·7	G
Roche Bois	Port Louis ...	Dec. 42									+								19·5	nil
Tamarin Bay	Black River ...	Feb. 44													+				19·1	G
Pte. d'Esny	Grand Port ...	Jan. 44													+				18·0	nil
Grand'Baie	R/Rempart ...	July 43												+					14·9	C, G
Roche Bois	Port Louis ...	Aug. 42											+						13·4	G
Bain des Dames	Port Louis ...	Dec. 42														+			13·0	nil
Roche Bois	Port Louis ...	Aug. 42											+						12·6	G
Peyrébère	R/Rempart ...	Mar. 44												+					12·3	G
Albion	Black River ...	Nov. 42																+	11·9	G
Bain des Dames	Port Louis ...	Dec. 42													+				10·5	nil
Albion	Black River ...	Oct. 42														+			10·2	G
Pte. d'Esny	Grand Port ...	Jan. 44							+										10·0	nil
Roche Bois	Port Louis ...	Oct. 42												+					9·9	G
Bain des Dames	Port Louis ...	Dec. 42														+			8·0	nil
Tamarin Bay	Black River ...	Feb. 43									+								8·0	G
Roche Bois	Port Louis ...	Aug. 42														+			7·6	G
Roche Bois	Port Louis ...	Aug. 42			+														7·3	G
Sable Noir	Port Louis ...	Apr. 43												+					7·3	C, G, F
Pte. d'Esny	Grand Port ...	Oct. 43						+											7·3	nil
Pte. d'Esny	Grand Port ...	Oct. 43		+															6·6	nil
Tamarin Bay	Black River ...	Apr. 43													+				6·2	G

*G=*A. gambiae* C=*A. coustani* F=*A. funestus* Mac=*A. maculipalpis*

TABLE VI—*continued.*

Range of pH+Salinity in Natural Breeding Places.

Locality	District	Date	7·0	7·2	7·3	7·4	7·5	7·6	7·7	7·8	7·9	8·0	8·1	8·2	8·3	8·4	8·5	8·6	Chlorides grs. NaCl per litre	Species of *Anopheles**
Tamarin Bay	Black River ...	Mar. 43														+			6·1	G
Albion	Black River ...	Nov. 43										+							5·2	G
Trou aux Biches	Pamplemousses ...	Aug. 43						+											5·0	G
Le Chaland	Grand Port ...	July 42			+														4·8	nil
Trou aux Biches	Pamplemousses ...	Oct. 42														+			4·5	G
Roche Bois	Port Louis ...	Aug. 42			+														4·4	G
Pte. aux Sables	Black River ...	July 43												+					4·0	F
Bain des Dames	Port Louis ...	Mar. 43													+				4·0	G
Pte. aux Sables	Black River ...	July 43												+					4·0	F
Choisy	Pamplemousses ...	Aug. 43													+				3·9	G
Bain des Dames	Port Louis ...	Sept. 42									+								3·7	G
Pte. aux Sables	Black River ...	July 43										+							3·5	C, G
Grand'Baie	R/Rempart ...	May 43							+										3·4	nil
Bain des Dames	Port Louis ...	Sept. 42						+											3·2	G
Solitude	Pamplemousses ...	Mar. 44										+							2·9	G
Pte. aux Sables	Black River ...	July 43								+									2·4	nil
Grand'Baie	R/Rempart ...	Aug. 43								+									2·2	nil
Sable Noir	Port Louis ...	Apr. 43													+				2·0	C, F
Mare Voundre		May 43	+																1·9	nil
Sable Noir	Port Louis ...	Apr. 43										+							1·8	C, F
Trou aux Biches	Pamplemousses ...	Aug. 43												+					1·7	nil
Pte. aux Sables	Black River ...	Oct. 42														+			1·3	G, F
Bain des Dames	Port Louis ...	Sept. 42													+				0·9	G
Sable Noir	Port Louis ...	Oct. 42																+	0·9	G
Riche Mare	Flacq	Mar. 43								+									0·8	C, G
Yemen	Black River ...	Apr. 43					+												0·8	C, G, Mac.
La Caverne (Vacoas) ...	Pl. Wilhems ...	Jan. 44					+												0·7	nil
Roche Bois	Port Louis ...	Mar. 43														+			0·7	C, G
Arsenal	Pamplemousses ...	Jan. 44						+											0·5	C, G
River Papayes	Black River ...	Feb. 44											+						0·4	G
Case Noyale	Black River ...	Aug. 43							+										0·4	C, G, F, Mac.
Trou aux Biches	Pamplemousses ...	Aug. 43										+							0·4	nil

*G=*A. gambiae* C=*A. coustani* F=*A. funestus* Mac=*A. maculipalpis*

TABLE VI—*continued.*

Range of pH+Salinity in Natural Breeding Places.

Locality	District	Date	7·0	7·2	7·3	7·4	7·5	7·6	7·7	7·8	7·9	8·0	8·1	8·2	8·3	8·4	8·5	8·6	Chlorides grs. NaCl per litre	Species of *Anopheles**
Roche Bois	Port Louis ...	Aug. 42								+									0·3	C
River Papayes	Black River ...	Aug. 42						+											0·3	C, F
Yemen	Black River ...	Aug. 42											+						0·3	C
Tombeau Bay	Pamplemousses ...	Oct. 42													+				0·3	C, F
River Papayes	Black River ...	Oct. 42						+											0·3	C
Sable Noir	Port Louis ...	Nov. 42											+						0·3	F
Riv. la Chaux	Grand Port ...	Aug. 42								+									0·2	F
Arsenal	Pamplemousses ...	Oct. 42													+				0·2	C, F
Roche Bois	Port Louis ...	Jan. 43					+												0·2	F
River Papayes	Black River ...	Feb. 43								+									0·2	C, G
Yemen	Black River ...	Feb. 43							+										0·2	C, G, Mac
River Papayes	Black River ...	Apr. 43								+									0·2	C, G
Yemen	Black River ...	Apr. 43																	0·2	C, G, Mac
Roche Bois	Port Louis ...	Apr. 43								+									0·2	F
Tombeau Bay	Pamplemousses ...	July 43									+								0·2	nil
Cressonville	Black River ...	Feb. 44											+						0·2	C, G, F
Mare Dodo	Grand Port ...	Aug. 42					+												0·1	C, F
River Papayes	Black River ...	Nov. 42						+											0·1	C
Cressonville	Black River ...	Feb. 43					+												0·1	C, F
Alma	Moka	Feb. 43				+													0·1	C, F
Bambou	Black River ...	Mar. 43					+												0·1	F
River Papayes	Black River ...	Apr. 43					+												0·1	C, G, F
Cressonville	Black River ...	Apr. 43							+										0·1	C, G, F
Pailles	Moka	Apr. 43						+											0·1	C, G, F
Pailles	Moka	Apr. 43														+			0·1	C, F
Forest Side	Pl. Wilhems ...	Apr. 43	+																0·1	C
Sable Noir	Port Louis ...	July 43								+									0·1	nil
La Caverne (Vacoas) ...	Pl. Wilhems ...	Jan. 43	+																0·1	nil
Pte. aux Sables	Black River ...	Feb. 44											+						0·1	G, F
Solitude	Pamplemousses ...	Mar. 44											+						0·1	nil
Calvert Moonooa (Tenre Rouge)	Pamplemousses ...	Mar. 44												+					0·1	C, F

*G=*A. gambiae* C=*A. coustani* F=*A. funestus* Mac=*A. maculipalpis*

Tolerance of A. gambiae *to Saline Waters.*

In coastal swamps in which the water may contain sodium chloride in proportions varying from 0·1 grm. to 23 grams per litre (2/3 pure sea water), the pH is generally between 7·5 and 8·4, and *A. gambiae* appears to flourish equally well at both extremes. The species can be made to pass through several stages in pure sea water, and in one salt pan, larvae were found in a slightly viscous salt solution rated at 65 grms. NaCl per litre. Gebert (1936) has recorded that *A. gambiae* may complete its development in water of very high NaCl content. Such conditions are believed to be produced by drying out of breeding places of lesser initial salinity, and no emergence has been observed in waters exceeding 50 grms. per litre. The Culicine, *Culex tritaeniorhynchus*, Giles, captured in the same saltpan, emerged successfully at 65 grms. NaCl per litre. In nature, the limits of tolerance are well covered by marshes supporting a growth of Voundre (*Typha gavanica*), Fougere (*Acrostichum aureum*) or Herbe La Mare (*Paspalum vaginatum*), but are usually exceeded by pure stands of mangrove. However, on the proximal edges of such swamps, and especially where seepages or storm water channels join the swamp, conditions become favourable to *A. gambiae*; it abounds in sunny shallow pools amongst the mangroves, where salinity may fall to 20 per cent. and inundation only takes place at the highest spring tides. It has thus been the policy to clear a coastward strip of any mangrove swamp found within 1 mile of the protected sites, and to investigate carefully such parts of the swamps as are not flushed with sea water at each normal high tide.

Tolerance of A. funestus *to Saline Waters.*

It can be seen from Table VI that *A. funestus* enters the list at an upper limit of 7·3 parts NaCl per thousand, but is of regular occurrence only at salinities of 2 per cent. and below.

In laboratory tests, series of ten 4th stage larvae of *A. funestus* were reared in bowls of sea water diluted with tap water to varying degrees of salinity. The results are tabulated below :—

TABLE VII.

Day	Parts NaCl per 1,000								Control natural water 1 per cent. NaCl
	1·5	3·0	5·0	6·0	7·0	8·0	9·0	10·0	
	Numbers of larvae alive								
1	10	10	10	10	10	10	10	10	10
2	10	10	10	9	8	1	5	—	10
3	10	10	10	5	5	1	—	—	10
4	10	7	8	5	3	—	—	—	8
5	7	7	7	5	3	—	—	—	8 (incl. 2 pupae)
6	6	7	7	4	—	—	—	—	7 (incl. 2 adults)
7	5+1 pupa	6	2		—	—	—	—	
8	2+4 pupae	6	—		—	—			
9	1 adult + 4 pupae	6	—		—	—			
10	5 adults	6	—		—	—			
11	—	3+3 pupae	—						
12	—	1+5 pupae	—						
13	—	4 adults	—						
14	—	—	—						
15	—	—	—						
			no adults		no adults		no adults		

It should be noted that the tests between 1·5 and 5·0 per cent. were made in October 1943 (mean temp. 22°) and the remainder under summer conditions (January 1944) at a mean temperature of 25°. Although far from complete, these laboratory findings agree tolerably well with the field observations.

In practice this means that *A. funestus* is unlikely to occur in coastal marshes that have frequent direct connection with the sea or, as in many cases, a constant small underground connection that causes an admittance of salt water to a degree depending on the seasonal state of the marsh. This species has not yet been found in conjunction with mangrove swamps or of the plant associations that occupy the transition zones to dry land (*Paspalum vaginatum*, *Acrostichum aureum*, etc.)

A. gambiae *in disused Saltpans.*

Saltpans are of common occurrence on the West coast where the rainfall is 30 to 35 inches. Normally, *A. gambiae* is absent, or, if present, is existing under most unfavourable conditions, specimens having been found in up to 65 grms. NaCl per litre. In the case of disused saltpans, however, where the regular flow of sea water is stopped, the salinity may fall during the wet season to a point below the normal limit of tolerance of *A. gambiae*. A good example has been treated during the present

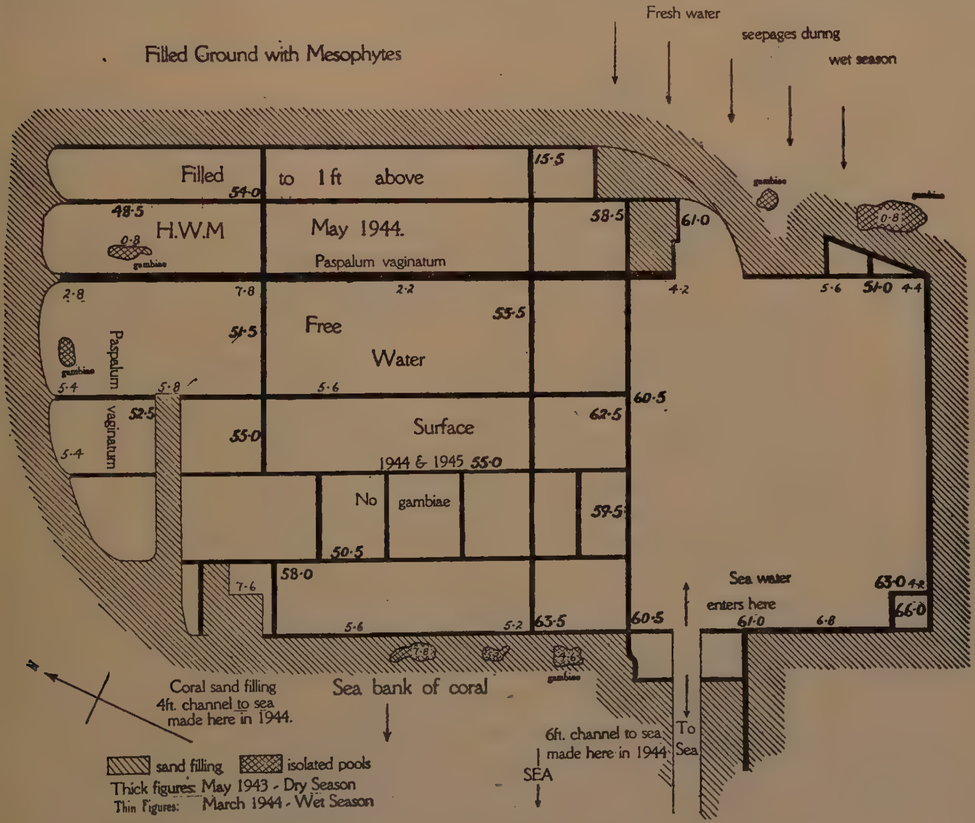
Fresh water
seepages during
wet season
Filled Ground with Mesophytes
Filled to 1 ft above
H.W.M
May 1944.
Paspalum vaginatum
Free Water Surface
1944 & 1945
No gambiae
Paspalum vaginatum
gambiae
54·0
48·5
0·8
15·5
58·5
61·0
7·8
2·8
2·2
55·5
54·5
5·4
5·8
5·6
52·5
55·0
5·4
55·0
4·2
5·6
51·0
4·4
60·5
62·5
59·5
50·5
58·0
7·6
5·6
5·2
63·5
60·5
Sea water
enters here
61·0
6·8
63·0
4·2
66·0
7·8
5·2
4·6
0·8
N
Coral sand filling
4ft. channel to sea
made here in 1944.
Sea bank of coral
6ft. channel to sea
made here in 1944
To Sea
SEA
sand filling
isolated pools
Thick figures: May 1943 - Dry Season
Thin Figures: March 1944 - Wet Season

Fig. 6. Plan of Cassis Saltpans (Diagrammatic).

investigation in the Cassis Saltpan, south-west of Port Louis harbour. The upper side of the pan, derelict since 1925, was overgrown with a pure stand of *Paspalum vaginatum* whilst the lower and main portion to seaward was quite free of vegetation and contained a firm highly saline surface of puddled clay.

The salinity at 22 points taken at random at the beginning of the dry season (May 1943) and again at 29 points taken at the wettest time of the year (March 1944) are shown in the sketch plan of the saltpan (fig. 6).

As regards *gambiae*, Ross (1908) recorded that only *Culex* was found in the pan which was then working. In 1943 only rare *Culex* were found in the saltpan proper (salinity about 50 per cent.) but, in holes surrounded by *Paspalum*, *Culex* was very abundant and *A. gambiae* were present in the proportion of about 1 per 1,000 *Culex* larvae. In seepage pools on the edges of the upper part of the pan (salinity 15·5 in 1943) *A. gambiae* was abundant and persistent. In 1943 the whole bed of the saltpan was unfortunately forked over and the high salinities of 50 to 60 per cent. disappeared for ever. In the wet period of March 1944 the general salinity was in the region of 4 to 7 per cent., but *A. gambiae* was confined to shallow pools on the edges, where wave action was minimal and the growth of *Spirogyra* favoured. In such pools,

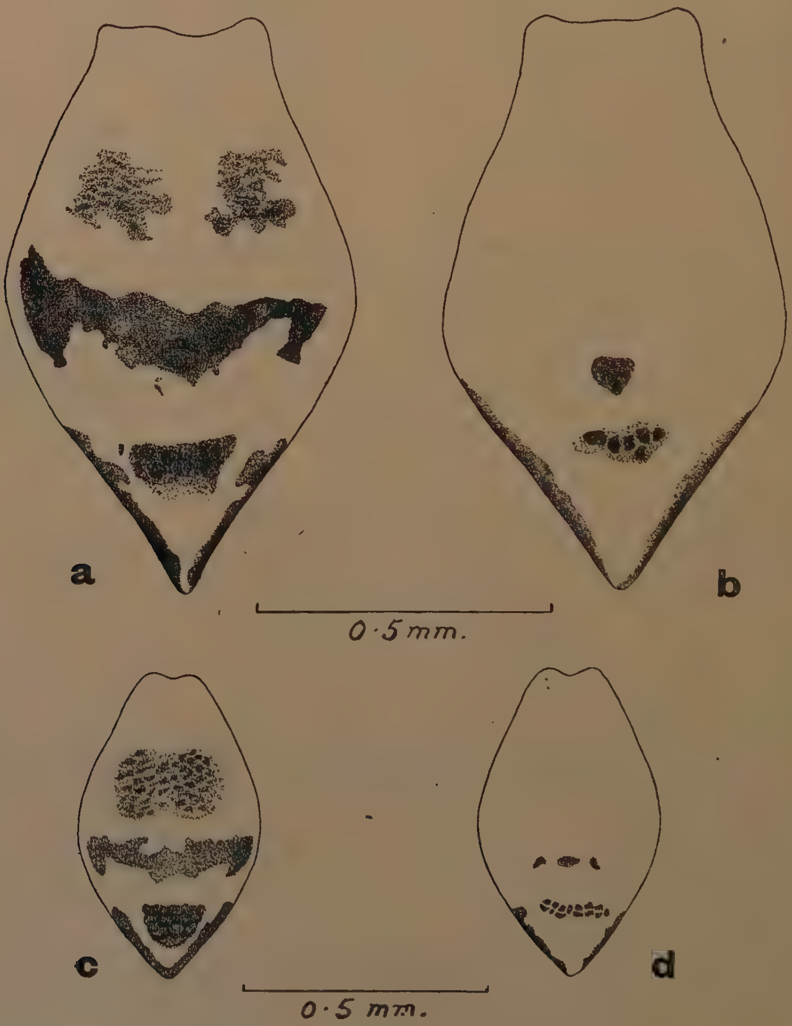

Fig. 7. Pigmentation of the fronto–clypeus of *Anopheles gambiae*, Giles. (*a*) Melanic from Roche Bois, Mauritius; (*b*) Normal from Roche Bois, Mauritius; (*c*) Melanic from Uganda, after Gibbins; (*d*) Normal from Uganda, after Gibbins.

especially when fed by seepage water from higher ground to the east, the salinity fell as low as 0·8 grm. NaCl per litre. This has been remedied by filling the edges of the pan with 1 to 2 ft. of coral sand, creating straight stone pitched edges where possible to promote wave action, and by opening two 6 ft. channels to the sea a few yards distant across a bank of loose coral. The salinity of the main pan is now constant at about 30 per cent., and it only remains to increase the level of the filling at a number of places where it has sunk and given rise to patches of *Paspalum*. As soon as this grass has yielded to a pure stand of *Chloris barbata* and *Dactyloctenum aegyptum* (herbe patte de poule), the work may be judged sufficient to prevent the formation of *gambiae* breeding places.

5. *Melanism in* A. gambiae *Larvae and Salinity*.

Melanism amongst the larvae of *A. gambiae* was found in nearly all breeding centres in the coastal belt ; the highest level at which melanic larvae were observed was about 500 to 600 ft. above sea level (Yemen, Black River). On examination, larvae of dark appearance were found to exhibit a typical pigmentation of the fronto-clypeus, closely resembling the melanic larvae described by Gibbins (1936) from Uganda (fig. 7). As far as is known at present, all melanic larvae give typical *gambiae* adults. In no case was 100 per cent. melanism found in one and the same breeding place. Observations to test whether melanism is correlated with high salinity or with pH are detailed below.

TABLE VIII.

Locality	Date	Occurrence of *gambiae*	Percentage melanism	Salinity, grams per litre	pH
Tamarin Bay	9.11.43	+++	70·0	25·7	8·1
Montee des Hussards	23.7.43	++	30·0	19·8	8·3
Tamarin Bay	4.3.44	+++	85·0	19·1	8·3
Pte. d'Esny	31.1.44	++	85·0	15·0	8·3
Grand'Baie	27.7.43	++	50·0	14·9	8·2
Roche Bois	9.11.42	+++	14·3	12·3	8·3
Albion	9.11.42	+++	58·6	10·4	8·4
Montee des Hussauds	19.1.43	+++	85·0	9·9	8·3
Tamarin Bay	13.3.43	+++	60·0	7·2	7·5
Roche Bois	24.3.43	+++	60·0	6·1	8·4
Albion	8.11.43	+++	85·0	5·2	8·0
Yemen	13.4.43	++	50·0	0·8	7·5
Tamarin Bay	16.3.43	+++	5·0	0·7	8·4
Cassis	23.3.44	++	70·0	0·6	8·0
River Papayes	2.2.44	+++	85·0	0·4	8·1
River Papayes	7.4.43	+++	80·0	0·2	7·8
Yemen	7.4.43	++	55·0	0·2	7·8

It will be seen that neither salinity nor pH appeared to have any particular influence on the occurrence of melanism in larvae of *A. gambiae*. It has been observed that in samples with very high percentages of melanism the water temperature was very high, around 40°C. Further controlled observations are needed. There is no evidence that abundance or lack of food is a factor in producing melanism. As previously remarked, *A. gambiae* var. *melas* has not been detected in Mauritius.

6. *Light.*

The importance of direct sunlight for optimum development of *A. gambiae* is too well known to need further confirmation, and control by shading the banks of streams and ponds has been practised in Mauritius on a limited scale for many years. The extent to which *A. funestus* will thrive under extremes of light intensity is a problem of the highest practical importance, but one that is complicated by the closely associated factors of temperature and food supply.

Preliminary observations in the laboratory under, at best, highly artificial conditions, gave somewhat conflicting results. Series of bowls each containing from 20 to 25 larvae of *A. funestus* were exposed to conditions varying from full laboratory light (200 to 450 ft. candles) to complete darkness. The following results were obtained at the Reduit Laboratory in February 1944 using 20 larvae at a mean laboratory temperature of 25·6°C. The larvae were obtained from Cressonville (900 ft.) and were fed on crushed *Spirogyra* and powdered dog biscuit.

TABLE IX.

Day	Number of larvae surviving	
	Light (diffused sunlight) 200–450 ft. candles	Darkness 0 ft. candles
1	20 larvae	19 larvae
2	20 ,,	18 ,,
3	20 ,,	16 ,,
4	20 ,,	15 ,,
5	20 ,,	15 ,,
6	20 ,,	12 ,,
7	20 ,,	12 ,,
8	20 ,,	10 ,,
9	6 pupae+14 larvae	10 ,,
10	6 ,, +14 ,,	10 ,,
11	6 ,, +14 ,,	9 ,,
12	6 adults+14 ,,	9 ,,
13	9 larvae	8 ,,
14	7 ,,	7 ,,
15	6 ,,	6 ,,
16	5 ,,	4 ,,
18	4 ,,	0 ,,
		no adults

Mean temperature 27°C. 24°C.

In other experiments, carried out with the same number of larvae, survival was generally greater in the bowls exposed to light, and adults were produced. When the bowls were wrapped in black cloth or shut in a dark cupboard, the larvae gradually died out and no adults were formed.

Measurements of light intensity in various situations including known breeding places of *A. funestus* were made with a sensitive photo-electric cell and a galvanometer reading in foot-candles.

TABLE X.

Place	Conditions	Time	Light Intensity (ft. candles)
1. Grand'Baie (Peyrébère)	Water surface on open marsh in brilliant sunlight	12.00	900
2. Reduit Laboratory ...	Bright sun no clouds	11.30	300
3. ,, ,, ...	Cloudy day	11.30	170
4. Alma	Gaps in jamrosa shade on stream bank	12.00	160
5. ,, ...	Full shade of jamrosa on sunny day	11.30	40
6. ,, ...	,, ,, ,, ,,	10.00	13
7. Reduit	Breeding cage in laboratory ...	11.30	10
8. Alma	Weedy edge of small stream ...	11.30	5
9. Solitude	Under " songe " leaves in marsh ...	11.30	0–5
10. Montagne Blanche ...	Under dense tree shade along small stream	09.30	2–5

Of the above habitats, *funestus* has been found in every one except the first four, one of the most favourable breeding places being beds in watercress and songe, where the larvae are rarely exposed to more than 5 to 10 ft. candles. At Montee des Hussards, 1 mile south of Port Louis, an abundant source of *A. funestus* was a mango orchard intersected with neglected irrigation channels. Although under a dense shade of the order 0 to 5 ft. candles, thousands of larvae were found by disturbing the layers of dead leaves blocking the channels. It is thought that the chief factor controlling *funestus* breeding under natural conditions is temperature and food supply. The temperature in a breeding place favourable to both vector species was measured repeatedly in different situations. The following represents a typical finding :—

TABLE XI.

Place	Conditions	Air Temp.	Water Temp.	Anopheles
Cressonville (600 ft.) ...	Open surface of green alga on still pool	25·8	33·5	gambiae ++++
,, ,,	Do. on brown alga	25·8	36·8	,, ++++
,, ,,	Surface under deep shade of " songe "	25·8	25·0	funestus +
,, ,,	4 inches deep near roots	25·8	24·0	,, +
Tamarin Bay (sea level)	Rockholes in River Papayes ...	32·0	39·0	gambiae ++++
,, ,,	Roots of water plant in light tree shade	32·0	31·5	funestus +

The temperature is thus about 7° to 8°C. lower in *A. funestus* than in *A. gambiae* breeding places. An interesting example of the control of *A. funestus* was provided in the present work by the clearing of reeds (*Typha javanica*) from a shallow swamp of 13·5 acres extent (Peyrébère, Grand'Baie). The water temperature was thereby raised to nearly 40°C. although light shade from the regrowth of reeds existed. *A. gambiae*, of course, populated the cleared marsh, but its flight habit appears to be more restricted than that of *A. funestus* and no further measures were attempted, since the marsh was over 2 miles from the protected area.

The practical recommendation arising from these observations, incomplete though they may be, is that whilst it is highly desirable, tree shade over streams should not be solely relied upon in the sluggish lowland watercourses. Streams and channels in which *A. funestus* is present should be regraded and rough trained, followed by the establishment of trees or bushes for anti-erosion purposes and for shading to eliminate *A. gambiae*. It does not appear probable that the light intensity could be reduced below the level of tolerance of *A. funestus* on the borders of large streams, especially as the foliage and branches are exposed annually to the depredations of cyclones just before the critical breeding season (March and April). Clean cut banks and increased water velocity must be aimed at in the lowlands, a view that was expressed by MacGregor (1924).

The best means of producing shade of maximum density along streams in regions up to 500 ft. above the sea, and especially in estuarine conditions with the water table near the surface, has been found in the leguminous tree, *Pongamia glabra* (*coculus*). This spreading, beech-leaved species grows well from seed, is hardy and easy to establish and is useless for any economic purpose. Furthermore, it is not relished by the ubiquitous goat, which has successfully frustrated a number of efforts with more palatable species. In reclaimed marshes or in low places where water may collect, close-planted *Casuarina* is recommended to provide shade and to create a carpet of needles inimical to Anopheline breeding. *Terminalia arjuna* and *Eucalyptus* are used as anti-erosion species, the former grows well in Grand Port District but suffers severe defoliation at the end of summer from a weevil (*Cratopus* sp.).

7. *Wind.*

The chief effects of wind as an environmental factor are :—

i. The normal dispersion of adult females from breeding to feeding sites is affected.

ii. Cyclonic winds destroy the adult insects.

i. *Flight Range and Wind.*

Since the nights in the summer season are frequently still, wind is not considered to play an important part in dispersing the adults. The flight range of Anophelines in Mauritius appears to be limited, perhaps because of relatively low night temperatures. It has been possible to leave uncontrolled a fairly extensive breeding place of *A. funestus* yielding 5 to 20 adults per room in a native village without a single *A. funestus* having been recorded over a period of 2 years from a service post 1·3 miles distant, across sugar cane fields. In other areas, the flight range of *A. gambiae* does not appear to exceed 1 mile and in the town of Port Louis it has been shown that houses ¼ of a mile from extensive breeding places are quite free of adults although numbers up to 150 per room can be found in the breeding area (Cassis).

An experiment on the usual lines was carried out to find out whether *A. gambiae* could be carried across a bay 1 mile wide in a direction at right angles to the wind. Over 2,000 adult *A. gambiae* bearing traces of methylene blue were released, but no trace of any Anopheline was found on large adhesive boards across the bay, although a mass breeding place 13·5 acres in extent was the normal source of the insects in the area of release.

In general, it has been taken as a working rule in the Malaria Unit that large breeding places 2 miles to windward and 1¼ miles to leeward should receive attention. It is regarded as an advantage if the breeding place is associated with a native village, provided that the village is at least 1 mile from the protected area.

ii. *Effect of the Cyclone of 15–17th January, 1945.*

It is sufficient to report here that the 100 m.p.h. winds of January 15th–17th caused an abrupt drop to zero in all check areas of Anopheline catches. The figure remained at zero for about 14 days, but within 28 days it was back to normal. The same observation was made in regard to larvae, except that the larval population increased more rapidly after the initial 14-day period.

Infectivity of Adults.

In order to obtain some information on the degree of infection in Anophelines in the Island, two highly infected regions were selected for investigation, viz., Pailles and Albion. Collection of live adults were made in huts from 8 a.m. to 3 p.m. Each insect, as soon as collected, was placed in a specimen tube 6 in. × 1 in., fitted with about ½ in. of damp cotton wool at the bottom and with organdi at the top. In the laboratory, the insect was fed on honey and dried raisins placed on the organdi —the honey was kept moist by means of a small wick dipped in a tube containing distilled water, as shown in the figure.

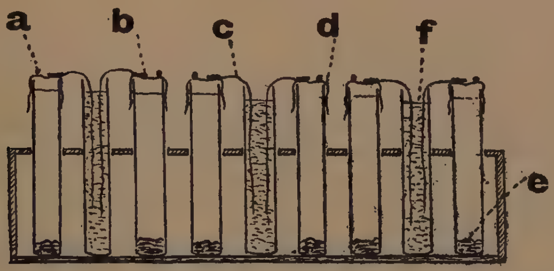

Fig. 8. (*a*), dried raisin ; (*b*), cotton wool with honey ; (*c*), wick ; (*d*), organdi ; (*e*), damp wool ; (*f*), water.

As over 90 per cent. of the insects collected were found to have taken a blood meal, they were usually dissected about 6 to 10 days after their capture, *i.e.* after digestion of the blood and development of oöcysts and sporozoites in their body. In one case a mosquito was found to be infected 30 days after being captured. In view of the controversy on the differentiation of oöcysts, etc., of the plasmodium, no attempt was made after dissection to determine the different species of the malaria parasites.

The results are given in Table XII.

A total of 1,140 mosquitos were dissected during the period October 1942 to March 1944. The number infected appears to be very high for so small a number dissected, the average infection being 3 per cent. It is worth recording that, on one occasion, out of six mosquitos collected in the same room, three were found to be infected. As previously mentioned, the species dissected was mainly *funestus* (over 95 per cent. of total insects dissected). As most of the specimens were collected in the same locality and by the same collectors, it can perhaps be considered that the figures represent the average infection rate for at least these two localities.

TABLE XII.

	1942			1943					
	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June
No. of mosquitos dissected ...	37	229	154	75	65	80	93	94	60
No. of infected stomachs (oöcysts)	—	2	2	2	—	—	—	5	3
No. of infected salivary glands (sporozoites)	—	—	—	—	—	—	—	2	3
Total number of infected mosquitos	—	2	2	2	—	—	—	5	3
Per cent. infection	—	0·87	1·3	2·66	—	—	—	5·32	5·0

TABLE XII—*continued.*

	1943						1944		
	July	Aug.	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.
No. of mosquitos dissected ...	65	28	26	45	—	6	18	23	42
No. of infected stomachs (oöcysts)	2	—	3	2	—	—	2	—	3
No. of infected salivary glands (sporozoites)	5	1	1	—	—	—	5	1	3
Total number of infected mosquitos	5	1	3	2	—	—	5	1	4
Per cent. infection	7·69	3·59	11·53	4·4	—	—	27·7	4·35	11·9

Mean per cent. infection : 3·07.

Summary and Conclusions.

1. A sketch is given of the Malaria Control Organisation in Service areas in Mauritius with a summarised history of malaria work in the Island since the first epidemic in 1865.

2. Keys to Mauritian Anophelines and their larvae with notes on their field recognition characters are followed by data on the distribution of adult Anophelines in the Colony and their relation to malaria intensity in different areas. *A. melas* appears to be absent from coastal swamps.

3. The influence of a number of environmental factors is discussed. It is concluded that temperature is an important limiting factor generally in winter and perennially in the highlands (above 1,000 ft.), but that the flushing action of heavy rains and probably the precipitation of food supply by colloidal iron moving under high rainfall conditions (over 100 ins.) from ferrugineous lavas both play their part in the natural control of *A. funestus* and *A. gambiae.*

The behaviour of *A. gambiae* with respect to temperature is expressed by an area enclosed by two symmetrical catenary curves, that illustrate well the adverse influence on development of low temperatures normally occurring in winter on the coast and all the year round in the residential uplands above 1,000 ft. The thermal death point of *A. gambiae* larvae is about 42°C. and that of *A. funestus* 40°C. and the lower limit of larval activity is 16·5°C. *A. gambiae* develops most rapidly at an estimated temperature of about 37°C.

4. In the coastal regions *A. gambiae* possesses a marked tolerance to highly saline waters and is found commonly at the upper limit of mangrove (*Rhizophora mucronata*) where rainwater and seepages dilute the tidal water. *Paspalum vaginatum* swamp is a dangerous source of *A. gambiae*. The pH of natural breeding places varies from 7·0 to 8·6 and appears to have little influence in affecting distribution in the regions below 1,000 ft.

5. Melanism in *A. gambiae* larvae was found in nearly all lowland areas. It was unrelated to salinity but may be correlated with high temperatures.

6. A consideration of the effect of light on the breeding of Anophelines leads to the conclusion that shade alone is unlikely to be sufficient to control *A. funestus* in sluggish lowland waters. The apparent effect of shading is almost certainly due to the lowered temperature rather than to critical reductions in light intensity.

7. The flight range of Anophelines does not normally exceed 1 mile, and in the town of Port Louis, houses within ¼ mile of a large seasonal breeding area were quite free of both *A. gambiae* and *A. funestus*.

8. The cyclone of 1945 with wind velocity of nearly 100 miles per hour caused a complete disappearance of both adults and larvae for a period of 14 to 21 days.

9. The natural infectivity of *A. funestus* is in the order of 3 to 4 per cent. but locally, infections up to 25 per cent. have been recorded. Infection is highest at the end of the transmission period, *i.e.* June to September.

Acknowledgements.

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A SPRAYING APPARATUS AND TESTING CHAMBER FOR INVESTIGATING THE RESIDUAL ACTION OF INSECTICIDAL DEPOSITS.

By J. E. Webb.
The Cooper Technical Bureau, Berkhamsted.

(Plate I.)

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Introduction.

Recent developments in the use of residual deposits of persistent contact insecticides such as 2 : 2-bis (*para*chlorophenyl)-1 : 1 : 1-trichlorethane (DDT) and 1 : 2 : 3 : 4 : 5 : 6-hexachloro*cyclo*hexane (666) to control insect pests and parasites call for a method for evaluating and investigating the lasting properties of such deposits on a variety of surfaces under different conditions of weathering. The first consideration in this problem lies in the accurate control of the weight and distribution of the deposit to be tested. Barnes (1945 & 1945a), working with *Cimex lectularius*, exposed the insects to deposits of various insecticides laid on the surface of blocks (2 in. × 2½ in.) of some of the materials used in the construction of buildings. To obtain an even deposit of known weight she sprayed the blocks with solutions of the insecticides in a " Potter Tower " (Potter, 1941). This method, however, is unsuitable for testing insecticidal deposits against flying insects such as house-flies and mosquitos chiefly owing to the comparatively small area which can be treated with Potter's apparatus. In this paper a spraying tower is described in which panels up to 20 cm. square can be treated evenly with a known weight of insecticide. These panels can then be built up to form a chamber in which the residual effect of insecticidal deposits can be studied on a variety of test insects. The physical performance of the spraying tower has been investigated both with respect to petroleum distillate and to water sprays. The behaviour of these spray mists under different conditions of spraying supports many of the conclusions reached by David (1946).

Construction of the Spraying Tower.

The spraying tower (see Pl. 1, fig. 1; text figs. 1 & 2) consists of a 3 ft. length of asbestos-cement pipe, 11½ in. internal diameter, firmly fixed to a stout wooden base and surmounted by a wooden box approximately 1 ft. 6 in. cube internal measurements. On one side, the lower edge of the pipe is cut away to a height of 1 in. to allow a sliding wooden tray to be inserted. This tray carries the object to be sprayed and when in position should completely seal the slit-like aperture formed by the cut-away portion of the pipe. In practice it has been found difficult to obtain a perfectly air-tight seal although a close approximation to this ideal condition has been reached by lining the contacting surfaces between the pipe and the tray with strips of plasticine. On one side of the upper chamber there is a glass observation window. Beneath this window, extending almost the entire width of the chamber, there is a horizontal slit through which a sliding panel can be inserted.

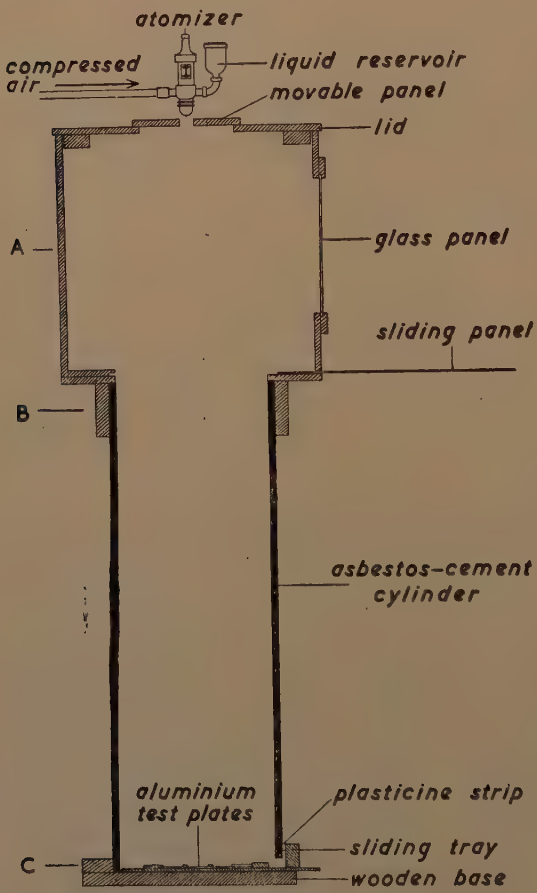
atomizer
compressed air
liquid reservoir
movable panel
lid
A
glass panel
sliding panel
B
asbestos-cement cylinder
aluminium test plates
plasticine strip
sliding tray
C
wooden base

Fig. 1.
The spraying tower seen in longitudinal section.

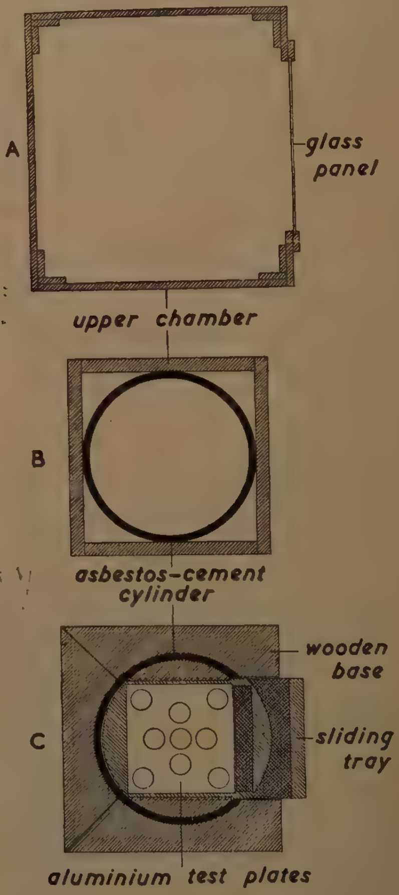
A
glass panel
upper chamber
B
asbestos-cement cylinder
wooden base
C
sliding tray
aluminium test plates
Scale
0
6
12 INCHES

Fig. 2.
The spraying tower seen in transverse section at the levels A, B and C indicated on fig. 1.

The panel runs in grooves in the floor of the upper chamber, completely separating it from the lower cylindrical portion of the tower. The upper chamber is fitted with a lid in the centre of which is an aperture 7 in. in diameter. This aperture is covered by a movable panel 8 in. square in the centre of which is a second aperture 1 in. in diameter. Immediately above this second aperture is the atomiser mounted on an adjustable arm. The atomiser used is an Aerostyle pneumatic spray gun, type "AD", fitted with a W/concentric head and a 0·5 mm. fluid tip. The compressed air is supplied by a ¼ h.p. compressor and passes through an air-line comprising two reducing valves, a sintered glass filter, a compressed air bottle and a piston-type master valve. Pressure gauges are fitted on the compressor, each reducing valve and the compressed air bottle. With this equipment the air flow is adequately stabilised and can be controlled up to pressures of 25 lb./sq. in. A lower limit of pressure is set by the gun itself, the spring-loaded needle-valve of which fails to open at pressures below 20 lb./sq. in.

Adjustment of the Atomiser.

The atomiser is adjusted to spray a given volume of liquid as rapidly as possible, firstly by unscrewing the nut controlling the needle-valve to allow the valve to open to the fullest extent, and secondly by altering the position of the air nozzle in relation to the fluid tip until atomisation is completed in the minimum time. When this position has been found the air nozzle is secured with a lock-nut. The air supply is then regulated to the desired pressure by means of the two reducing valves.

A sheet of paper is pasted on to the sliding panel separating the upper chamber from the lower portion of the tower and marked with diagonal lines so placed that, when the panel is pushed in, the point of intersection of the lines lies on the axis of the tower. The square panel covering the hole in the lid of the upper chamber is now replaced by a piece of card from which is suspended a plumb-bob. This is adjusted until the plumb-bob hangs directly over the point of intersection of the diagonals marked on the paper. The atomiser is centred with respect to the tower when it is vertically above the point of origin of the plumb-line. The card with plumb-bob is replaced by the panel with the 1 in. circular hole immediately beneath the atomiser. A small quantity of dye solution is sprayed on to the marked sheet of paper and the distribution of the deposit noted. If this deviates from the centre of the paper, correction is made by small adjustments in the position of the atomiser. When the atomiser has been centred the sliding panel is removed and the slit through which it is inserted is covered by a strip of adhesive tape.

Spraying Procedure.

Before spraying, the temperature of the room is controlled at 20°C. (approx.) and the air pressure, with the atomiser spraying, is adjusted to 25 lb./sq. in. A priming dose of 5 cc. of the insecticidal solution to be used is sprayed to wet the fluid duct of the atomiser. Where the insecticide is in a petroleum distillate base, this single priming dose is also sufficient to raise the petroleum distillate vapour content of the atmosphere of the tower so near to saturation point that evaporation of spray droplets during subsequent sprayings is negligible. With water base preparations, however, it is necessary to spray at least five priming doses of 5 cc. each before the humidity of the atmosphere of the tower is raised to the point where this condition obtains. Ten minutes after the last priming dose has been discharged, the sliding tray, with the object to be sprayed upon it, is inserted into the base of the tower and sprayed with 5 cc. of liquid, the valve controlling the air supply being closed immediately all the liquid has been delivered. The time taken to spray through is noted and should not vary more than a few tenths of a second from one spraying to the next. Five minutes after spraying the tray is removed, the sprayed object replaced by the next and the atomiser re-charged. After a further five minutes, *i.e.* ten minutes after spraying, the tray is inserted in the tower and the next spraying is carried out. It is important for replication of deposits throughout a series of similar tests that this rhythm of spraying should be accurately maintained.

Where it is required to deposit different weights of an insecticide on to a surface, this may be done by varying either the concentration of the insecticide in the liquid to be sprayed or the rate of delivery of the atomiser by adjustment of the needle-valve.

Determination of Weight of Deposit.

The estimation of deposits by weighing is a lengthy process and, in view of the small quantities involved, is of doubtful accuracy. Potter (1941) calculated the weights of deposits of insecticide from the total weight of spray collected, but David (1946) has shown conclusively that, owing to evaporation of spray droplets during their passage from the atomiser to the surface on which they are deposited, the concentration of insecticide in the liquid at that surface may in fact be considerably greater than the original concentration before spraying. The total weight of spray deposited, therefore, may bear little relationship to the weight of insecticide which can only be determined accurately by a direct method of estimation. For this purpose Potter and Hocking (1939) and later David (1946 & 1946a) incorporated dyes in the sprays they used and estimated the quantity of dye collected in a filter or on a surface by comparison with standard solutions of known dye content. The method suggested here is a refinement of the colorimetric method in so far as the personal element in making the comparisons between dye solutions of different strengths is eliminated by use of the Spekker Photo-electric Absorptiometer. Details of this instrument are given by Haywood and Wood (1944).

The physical performance of the spraying tower in so far as it affects the weights of deposit obtained under different conditions has been examined with respect to two solutions, firstly a 1 per cent. W/V solution of " Oil Golden Yellow 13777 "* in petroleum distillate, and secondly a 1 per cent. W/V solution of " Orange 60506 "* in distilled water. In each spraying test estimations are made of the weight of dye deposited on nine circles 4 cm. in diameter disposed about a 20 cm. square as shown in fig. 3. This is achieved in the following manner. Two stout aluminium plates each 20 cm. square are used; the first, which is entire, serves as a base-plate and the second, which is drilled with nine holes 4 cm. in diameter, is a test-plate. One of the holes is central and the remainder are arranged about it in two sets of four on circles of radius 5 cm. and 10 cm. respectively. The base-plate is placed on the sliding tray in such a position that when the tray is inserted into the tower the centre of the plate coincides with the axis of the tower. The base-plate is covered with a sheet of semi-absorbent paper on which the positions of the holes in the test-plate have been marked. Nine circles of No. 1 Whatman filter paper 5·5 cm. in diameter are placed in these positions and numbered from 1–9. The test-plate is then placed over the filter papers so that on each paper there is exposed a circular area 4 cm. in diameter. The tray and test-papers are then inserted into the tower and sprayed in the manner described in the previous section. On removal from the tower five minutes after spraying the test-plate is cleaned with an absorbent rag and the papers each bearing a circular area of dye deposit are placed into boiling tubes containing 50 cc. of either petroleum distillate or distilled water according to the liquid sprayed. The tubes are then corked and the papers left in the solvent until all the dye is in solution. Fresh papers are laid out on the base-plate and covered with the test-plate in readiness for the next spraying. The time of ten minutes between sprayings, *i.e.* five minutes between removal of one batch of sprayed test papers and the insertion of the next batch, was chosen as the minimum time in which one operator could comfortably perform these tasks.

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* Obtained from Messrs. Williams (Hounslow) Ltd.

When the dye has been leached from the test-papers, the solution in each boiling tube is stirred and the quantity of dye in solution estimated by means of the Spekker Photo-electric Absorptiometer. The Absorptiometer was calibrated with a wide range of dye solutions of known strength. The filters used were No. 601 for solutions of "Oil Golden Yellow 13777" and No. 603 for solutions of "Orange 60506". This method enables estimations of dye to be made to 0·01 mg. with great rapidity.

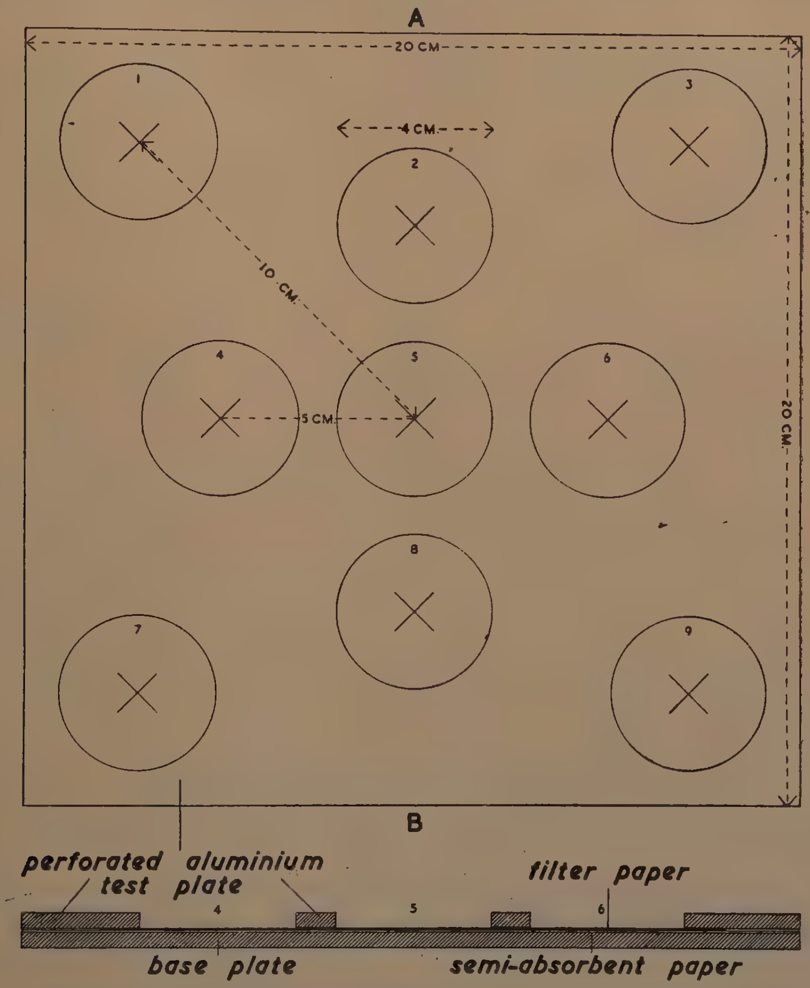

Fig. 3.—A. The arrangement of the holes drilled in the aluminium test-plate; B. The test and base-plates shown in section with the test-papers trapped between them

Replication and Distribution of Deposits.

The distribution and replication of the deposits of dye obtained with the spraying tower were tested for both the petroleum distillate and the aqueous dye solutions in series of tests carried out at 20°C. and 25 lb./sq. in. spraying pressure and repeated from day to day. Throughout these series of tests, the mean weight of dye deposited in each series was maintained within narrow limits. The Standard Deviation of the mean weight of deposit per test-paper for both spray liquids, both from one test to another and between different positions on the area sprayed in any one test, fell between ±0·8 per cent. and ±3·0 per cent. of the mean. There was, however, in both petroleum distillate and aqueous series a tendency for the deposits to be heavier in positions 7, 8 and 9 than in positions 1, 2 and 3. This was thought to be due to a slight leakage of air at the point of insertion of the tray causing a drift of spray droplets towards positions 7, 8 and 9. No doubt with an improved fitting of the tray these differences could be eliminated entirely.

Factors governing the Weight of Deposit.

1. *Degree of saturation of the atmosphere within the tower with vapour of the liquid to be sprayed.*

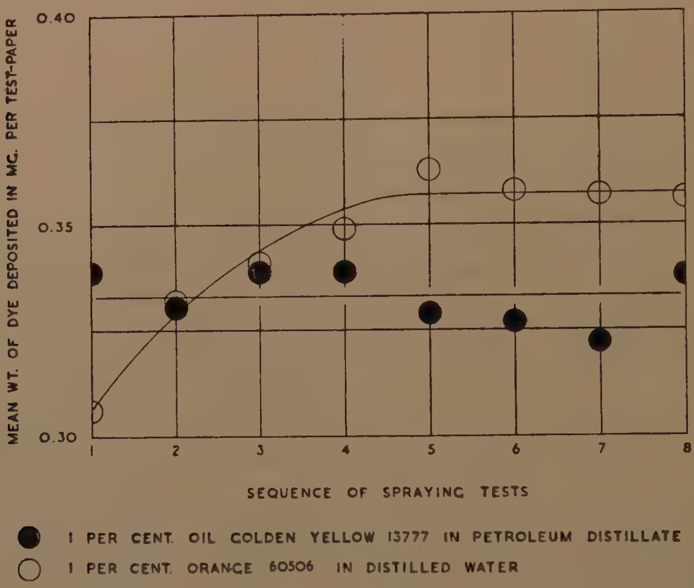

Fig. 4.—Graph showing the relationship between the weight of deposit and the number of consecutive spraying tests carried out.

The series of tests given in Tables I and II and fig. 4 indicate the number of priming doses of liquid which must be sprayed into the tower before replication of deposit can be obtained from either petroleum distillate or aqueous sprays. Whereas with a petroleum distillate dye solution replication of deposit was obtained after the initial priming dose used to wet the fluid duct of the atomiser had been sprayed (see Table I and fig. 4), with an aqueous dye solution the weight of deposit steadily increased with consecutive sprayings until the fifth test and then remained at a constant level (see Table II and fig. 4). It is necessary, therefore, to charge the atmosphere of the tower with five priming doses of 5 cc. each of an aqueous spray before replication of deposit can be obtained. This is undoubtedly due to the degree of saturation of the atmosphere within the tower with vapour of the liquid sprayed and its effect on the rate of evaporation of the spray droplets. At each successive spraying, the droplets will partially evaporate and increase the degree of saturation of the atmosphere of the tower with vapour. If the tray at the base of the tower were not withdrawn between sprayings, evaporation of droplets would become less and less at each spraying and finally cease when the vapour pressure within the tower reached saturation point. Thus as the rate of evaporation declined,

TABLE I.

5 cc. of 1 per cent. petroleum distillate solution of "Oil Golden Yellow 13777" sprayed at 25 lb./sq. in. in 12·7–13 secs. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings, 10 mins. One priming dose of 5 cc.

Position of Test-Paper	Weight of Dye deposited in mg. per Test-Paper								Mean
	Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	
1	0·33	0·33	0·33	0·33	0·33	0·32	0·32	0·32	0·326
2	0·34	0·33	0·33	0·34	0·33	0·33	0·32	0·33	0·331
3	0·35	0·34	0·33	0·34	0·33	0·33	0·33	0·32	0·334
4	0·33	0·32	0·34	0·34	0·33	0·32	0·32	0·33	0·329
5	0·33	0·33	0·34	0·34	0·33	0·32	0·32	0·35	0·333
6	0·35	0·35	0·35	0·34	0·33	0·34	0·33	0·35	0·343
7	0·33	0·32	0·34	0·34	0·32	0·32	0·32	0·34	0·329
8	0·34	0·32	0·35	0·34	0·33	0·32	0·32	0·35	0·334
9	0·35	0·34	0·34	0·34	0·33	0·34	0·32	0·35	0·339
Mean	0·339	0·331	0·339	0·339	0·329	0·327	0·322	0·338	0·333

Standard Deviation of Means at Positions 1–9 = ± 0·0053=1·6% of the mean.
Standard Deviation of Means of Tests 1–8=± 0·0067=2·0% of the mean.

TABLE II.

5 cc. of 1 per cent. aqueous solution of "Orange 60506" sprayed at 25 lb./sq. in. in 11·7–12·0 secs. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. One priming dose of 5 cc.

Position of Test-Paper	Weight of Dye deposited in mg. per Test-Paper								Mean of Tests 5–8
	Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	
1	0·29	0·31	0·34	0·34	0·35	0·37	0·34	0·36	0·355
2	0·30	0·32	0·35	0·35	0·35	0·35	0·35	0·36	0·353
3	0·32	0·35	0·35	0·35	0·37	0·34	0·36	0·37	0·360
4	0·30	0·33	0·34	0·35	0·36	0·36	0·35	0·35	0·355
5	0·30	0·32	0·33	0·34	0·35	0·36	0·37	0·35	0·358
6	0·31	0·35	0·34	0·34	0·37	0·34	0·35	0·36	0·355
7	0·31	0·33	0·34	0·37	0·39	0·37	0·35	0·35	0·365
8	0·30	0·33	0·33	0·35	0·37	0·36	0·38	0·34	0·362
9	0·32	0·35	0·35	0·35	0·36	0·37	0·38	0·36	0·368
Mean	0·306	0·332	0·341	0·349	0·363	0·358	0·359	0·356	0·359

Standard Deviation of Means at Positions 1–9=± 0·0051=1·42% of the mean.
Standard Deviation of Means of Tests 5–8=± 0·0029=0·81% of the mean.

there would be a corresponding increase in the weight of deposit obtained at each spraying test, reaching and being maintained at a maximum weight when the atmosphere of the tower became saturated with vapour. The removal of the tray for five minutes between successive sprayings would lead to the replacement of some of the saturated air within the tower by unsaturated air from without and thus a fall in the vapour pressure within the tower would occur. In a series of sprayings a close replication of deposit from one spraying to the next would not obtain until an equilibrium was set up between the increase in vapour pressure within the tower due to spraying and the reduction due to the removal of the tray. At this point of equilibrium, which would vary according to the period for which the tray is withdrawn, there should be a constant vapour pressure within the tower at the time of spraying. The estimations of weight of deposits are, however, made on the non-volatile constituent of the spray so that evaporation of the spray droplet is of no importance provided that the droplet remains heavy enough to settle within the five minutes after spraying for which the test-papers are exposed. Only droplets which have evaporated beyond this point or have evaporated completely and leave a residue of dye too light to settle will affect the weight of deposit. A standard mist, therefore, exposed to a definite vapour pressure within the tower for a given time should produce a standard weight of deposit. The only way of ensuring that at the commencement of each spraying the percentage saturation of vapour is always the same is to adopt a standard rhythm of spraying and to spray as many priming charges of liquid as are necessary to bring the rise and fall of the vapour content of the atmosphere of the tower into equilibrium with the spraying rhythm. With petroleum distillate sprays this equilibrium seems to be reached with one priming dose but with aqueous sprays five priming doses are necessary. It is probable too, that with aqueous sprays the humidity of the atmosphere of the room will affect both the number of priming doses required and the percentage saturation of water vapour within the tower at which equilibrium is established, but as the humidity within the tower during spraying tests must approach near to saturation it is unlikely that small variations of humidity in the external air would make much difference to the weight of deposit, particularly if that humidity is low. Unfortunately in the present series of tests the humidity of the room in which the spraying tests were carried out was uncontrolled, although no discrepancies in the results appear to have arisen through neglect of this factor. It would be preferable, however, to control humidity where possible, particularly when aqueous sprays are used.

2. *Temperature.*

On theoretical grounds it would be expected that the weight of deposit obtained in any spraying test would vary with the temperature. An alteration in temperature by changing the physical characteristics, such as surface tension and viscosity, of the liquid to be sprayed would affect the nature of the mist produced and, by raising the vapour pressure of the liquid, would alter the rate and degree of evaporation of the spray droplets. To determine the influence of these factors on the weight of deposit, two series of tests were carried out with petroleum distillate and aqueous dye solutions, respectively, at 30°C. for comparison with the similar tests (see Tables I and II) carried out at 20°C. The results of these tests are given in Tables III and IV. It was found that with the petroleum distillate spray at 30°C. the mean weight of dye deposited was lower than that obtained at 20°C. and that the Standard Deviation of the weights of dye deposited on the test-papers at different positions was higher. Moreover, the weight of the deposit on those test-papers disposed towards the centre of the test-panel was greater than on those towards the periphery. This is in contrast to the more even distribution of dye obtained at 20°C. With the aqueous spray at 30°C., on the other hand, neither the mean weight of deposit nor the Standard Deviation of the weights of deposit on the test-papers at different positions was significantly different from those obtained at 20°C. It seems, therefore, that with aqueous sprays little is to be gained by a close control of the temperature at which spraying is carried out, or for that matter the humidity of the external atmosphere, for the raising of the temperature of the room to 30°C. must have reduced the relative humidity of the atmosphere considerably and yet no difference in the mean weight of deposit was observed. With the petroleum distillate spray, however, the reduction in weight of deposit following a rise of temperature from 20–30°C. shows that in this instance temperature should be controlled.

TABLE III.

5 cc. of 1 per cent. petroleum distillate solution of "Oil Golden Yellow 13777" sprayed at 25 lb./sq. in. in 11·5–11·8 secs. at 30°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. One priming dose of 5 cc.

Position of Test-Paper	Weight of Dye deposited in mg. per Test-Paper				Mean of Tests
	Test 1	Test 2	Test 3	Test 4	
1	0·28	0·26	0·27	0·30	0·278
2	0·30	0·27	0·29	0·30	0·290
3	0·30	0·26	0·28	0·30	0·285
4	0·31	0·28	0·28	0·30	0·293
5	0·31	0·28	0·29	0·30	0·295
6	0·30	0·29	0·28	0·30	0·293
7	0·31	0·29	0·28	0·30	0·295
8	0·31	0·29	0·29	0·30	0·298
9	0·30	0·28	0·28	0·29	0·288
Mean	0·302	0·277	0·282	0·299	0·290

Standard Deviation of Means at Positions 1–9 = ± 0·0061 = 2·1% of the mean.
Standard Deviation of Means of Tests 1–4 = ± 0·0124 = 4·3% of the mean.

TABLE IV.

5 cc. of 1 per cent. aqueous solution of "Orange 60506" sprayed at 25 lb./sq. in. in 11·0–11·9 secs. at 30°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. Five priming doses of 5 cc. each.

Position of Test-Paper	Weight of Dye deposited in mg. per Test-Paper				Mean of Tests
	Test 1	Test 2	Test 3	Test 4	
1	0·35	0·36	0·35	0·36	0·355
2	0·35	0·37	0·35	0·37	0·360
3	0·35	0·35	0·36	0·37	0·358
4	0·35	0·38	0·35	0·36	0·360
5	0·36	0·38	0·35	0·37	0·365
6	0·35	0·37	0·35	0·38	0·363
7	0·34	0·35	0·35	0·35	0·348
8	0·37	0·39	0·35	0·37	0·370
9	0·35	0·36	0·35	0·38	0·360
Mean	0·352	0·368	0·351	0·368	0·360

Standard Deviation of Means of Positions 1–9 = ± 0·0062 = 1·72% of the mean.
Standard Deviation of Means of Tests 1–4 = ± 0·0095 = 2·64 % of the mean.

It is probable, that a rise in temperature affects petroleum distillate sprays by altering the character or droplet size of the mist. This could occur in two ways, firstly by a reduction in viscosity and secondly by a reduction in surface tension. Either or both of these factors might cause an increase in the degree of atomisation with a rise in temperature, but, as they are also operative in the case of water sprays, perhaps some difference in the change of physical properties of the liquids with rise in temperature should be sought which is proportionately greater in petroleum distillate than in water. The viscosity in both liquids is reduced, but to approximately the same proportion, but surface tension is reduced to a proportionately greater extent in petroleum distillate than in water. This change in surface tension, therefore, may be responsible for a greater reduction of droplet size with rise of temperature when petroleum distillate is sprayed than obtains with water. Even if this is not so the smaller size of petroleum distillate droplets as compared with water droplets when the liquids are sprayed under similar conditions may mean that the droplet size in the former case is near some critical point, so that any further reduction in droplet size has an effect on their behaviour out of all proportion to that which would attend a similar reduction in size of the relatively coarse water droplets. The difference in size of droplets from petroleum distillate and water sprays may be directly attributed to the much lower surface tension of petroleum distillate as compared with water. Under the conditions of spraying adopted here, petroleum distillate droplets are still in suspension in the atmosphere of the tower five minutes after spraying, whereas with water this is not so to any marked extent, a fact which probably accounts for the lower weight of deposit obtained with petroleum distillate sprays at 20°C. than with water (see Tables I and II). An equal reduction in droplet size of both mists would result in a still higher proportion of the petroleum distillate droplets remaining in suspension in the atmosphere five minutes after spraying and a consequent reduction in weight of deposit, whereas the water droplets, although reduced in size, may still be deposited entirely within the first five minutes after spraying and thus no reduction in weight of deposit would follow.

3. *Spraying pressure.*

A further factor influencing droplet size in a spray mist and hence the weight of deposit obtained is the spraying pressure at which the liquid is atomised. A change in spraying pressure will mean an alteration in the amount of energy utilised during spraying and consequently in the degree of atomisation achieved. The greater the amount of energy used in breaking up the liquid into droplets the smaller the droplets obtained. To demonstrate this effect tests were made with both the petroleum distillate and the aqueous dye solutions at 20°C. and 22·5 and 20 lb./sq. in. spraying pressure for comparison with those carried out at 20°C. and 25 lb./sq. in. The results obtained are given in Tables V–VIII. It was found that, with the petroleum distillate dye solution the weight of deposit decreased with the spraying pressure (see Tables I, V and VI). With the aqueous spray, however, this result was not obtained: weights of deposit at a spraying pressure of 22·5 lb./sq. in. were not significantly different from those at 25 lb./sq. in., while at 20 lb./sq. in. the weight of deposit increased slightly (see Tables II, VII and VIII).

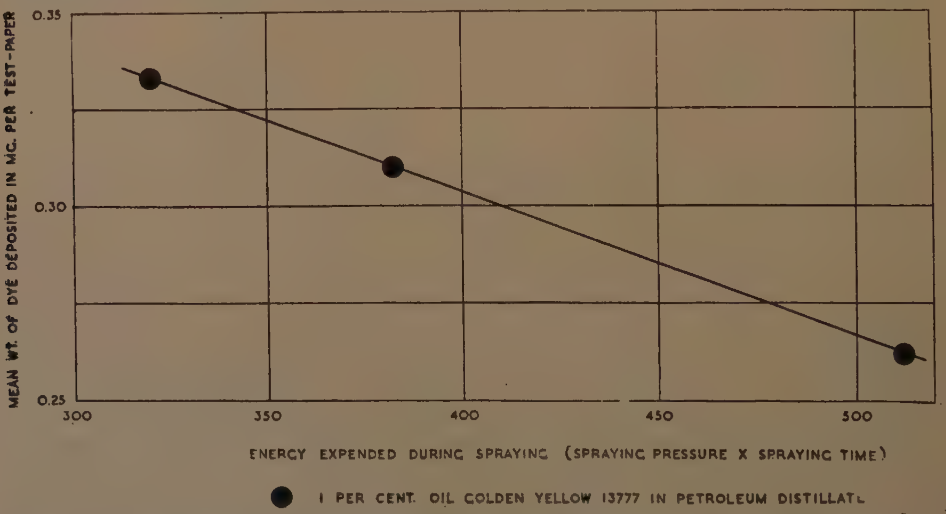

Fig. 5.—Graph showing the fall in weight of deposit obtained from petroleum distillate dye solution with increasing expenditure of energy during spraying.

TABLE V.

5 cc. of 1 per cent. petroleum distillate solution of " Oil Golden Yellow 13777 " sprayed a 22·5 lb./sq. in. in 16·5–17·5 secs. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. One priming dose of 5 cc.

Position of Test-Paper	Weight of Dye deposited in mg. per Test-Paper				Mean of Test
	Test 1	Test 2	Test 3	Test 4	
1	0·29	0·29	0·28	0·28	0·285
2	0·31	0·31	0·29	0·30	0·303
3	0·31	0·30	0·29	0·29	0·298
4	0·32	0·30	0·30	0·32	0·310
5	0·32	0·32	0·31	0·31	0·315
6	0·32	0·32	0·30	0·32	0·315
7	0·33	0·32	0·31	0·33	0·323
8	0·33	0·32	0·31	0·32	0·320
9	0·33	0·33	0·31	0·32	0·323
Mean	0·318	0·312	0·300	0·310	0·310

Standard Deviation of Means at Positions 1–9 = ± 0·0128 = 4·13% of the mean.
Standard Deviation of Means of Tests 1–4 = ± 0·0075 = 2·42% of the mean.

TABLE VI.

5 cc. of 1 per cent. petroleum distillate solution of " Oil Golden Yellow 13777 " sprayed at 20 lb./sq. in. in 25·3–26 secs. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between spraying 10 mins. One priming dose of 5 cc.

Position of Test-Paper	Weight of Dye deposited in mg. per Test-Paper				Mean of Test
	Test 1	Test 2	Test 3	Test 4	
1	0·26	0·25	0·25	0·25	0·253
2	0·26	0·26	0·25	0·27	0·260
3	0·25	0·26	0·24	0·27	0·255
4	0·25	0·26	0·25	0·27	0·258
5	0·27	0·26	0·25	0·27	0·263
6	0·27	0·26	0·25	0·27	0·263
7	0·28	0·27	0·27	0·28	0·275
8	0·28	0·27	0·26	0·28	0·273
9	0·27	0·28	0·24	0·26	0·263
Mean	0·266	0·263	0·251	0·269	0·262

Standard Deviation of Means at Positions 1–9 = ± 0·0074 = 2·82% of the mean.
Standard Deviation of Means of Tests 1–4 = ± 0·0079 = 3·02% of the mean.

TABLE VII.

5 cc. of 1 per cent. aqueous solution of "Orange 60506" sprayed at 22·5 lb./sq. in. in 12·9–13·5 secs. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. Five priming doses of 5 cc. each.

Position of Test-Paper	Weight of Dye deposited in mg. per Test-Paper				Mean of Test
	Test 1	Test 2	Test 3	Test 4	
1	0·37	0·34	0·35	0·36	0·355
2	0·36	0·35	0·35	0·35	0·353
3	0·35	0·35	0·35	0·34	0·348
4	0·36	0·34	0·35	0·36	0·358
5	0·35	0·35	0·37	0·35	0·355
6	0·35	0·35	0·38	0·35	0·358
7	0·36	0·34	0·36	0·36	0·355
8	0·35	0·35	0·38	0·36	0·360
9	0·34	0·35	0·36	0·35	0·350
Mean	0·354	0·347	0·361	0·353	0·354

Standard Deviation of Means at Positions 1–9 = ± 0·0039 = 1·1% of the mean.
Standard Deviation of Means of Tests 1–4 = ± 0·0057 = 1·61% of the mean.

TABLE VIII.

5 cc. of 1 per cent. aqueous solution of "Orange 60506" sprayed at 20 lb./sq. in. in 16·5–16·8 secs. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. Five priming doses of 5 cc. each.

Position of Test-Paper	Weight of Dye deposited in mg. per Test-Paper				Mean of Test
	Test 1	Test 2	Test 3	Test 4	
1	0·36	0·35	0·35	0·35	0·353
2	0·37	0·37	0·36	0·37	0·368
3	0·37	0·36	0·37	0·37	0·368
4	0·37	0·37	0·35	0·37	0·365
5	0·38	0·37	0·37	0·38	0·375
6	0·38	0·38	0·37	0·38	0·378
7	0·35	0·37	0·36	0·37	0·363
8	0·37	0·38	0·38	0·39	0·380
9	0·37	0·38	0·39	0·39	0·383
Mean	0·369	0·370	0·367	0·374	0·370

Standard Deviation of Means at Positions 1–9 = ±0·0095 = 2·57% of the mean.
Standard Deviation of Means of Tests 1–4 = ± 0·0029 = 0·8% of the mean.

TABLE IX.

	Petroleum Distillate			Distilled Water		
Spraying pressure in lb./sq. in. (P) ...	25	22·5	20	25	22·5	20
Spraying time in seconds (T) ...	12·8	17	25·6	11·8	13·2	16·6
Energy used in spraying (P × T) ...	320	382·5	512	295	297	332
Mean wt. of dye deposited in mg./Test-paper	0·333	0·310	0·262	0·358	0·354	0·370

These results are partly explained by a consideration of the different amounts of energy involved in the atomisation of the two liquids at different spraying pressures (see Table IX and fig. 5). The energy utilised in spraying, however, is not proportional to the spraying pressure, for as this pressure is reduced the time taken to spray through is increased to a varying amount according to the liquid sprayed. A figure proportional to the amount of energy used is given by the product of the spraying pressure and the time taken to spray the liquid. Thus it is seen from Table IX that, where petroleum distillate is sprayed, as the spraying pressure is reduced the amount of energy utilised is in fact increased owing to the greater time taken to spray through at the lower pressures. Moreover, the weight of deposit obtained from the petroleum distillate dye solution varied inversely with the energy expended during atomisation (see fig. 5). This can only mean that the weight of deposit in this instance varies directly with the droplet size of the mist and thus any factor affecting droplet size will also affect weight of deposit. It is, therefore, necessary to exercise a close control over all aspects of the spraying procedure where petroleum distillate is sprayed. It is seen from Table IX that, when the aqueous dye solution was sprayed, the quantity of energy involved at spraying pressures of 25 and 22·5 lb./sq. in. was substantially the same and at 20 lb./sq. in. only slightly greater. This would account for the lack of variation in the weights of deposit at the two higher spraying pressures although not for the slight rise in weight of deposit obtained at 20 lb./sq. in. It has already been suggested in the section on the effect of varying temperature that, as far as the petroleum distillate spray is concerned, a reduction in droplet size results in a reduction in weight of deposit merely because a greater proportion of the mist fails to settle in the time for which the test-papers are exposed in the tower, whereas with the aqueous spray it is probable that the droplets could suffer a reduction in size and still be large enough to settle within five minutes after spraying. This explanation would cover the fact that no reduction in weight of deposit was obtained with the aqueous spray at 20 lb./sq. in. spraying pressure : the slight increase in weight observed in this test may have been due to an increased turbulence caused by the longer spraying time.

The replication and distribution of the deposits obtained from petroleum distillate dye solution at 22·5 and 20 lb./sq. in. spraying pressure, as shown by the Standard Deviation of the mean weights at different positions and for different tests, respectively (see Tables V and VI), were less accurate than those obtained at 25 lb./sq. in. spraying pressure (see Table I) : a similar decrease in accuracy was observed when the weight of deposit was reduced by spraying at a higher temperature (see Table III). The distribution and replication of deposit from the aqueous spray at 22·5 and 20 lb./sq. in. spraying pressure was of the same order as that obtained at 25 lb./sq. in. (see Tables II, VII and VIII).

It is concluded that with aqueous sprays at 20°C., differences of spraying pressure of the order tested have little effect on the weight of deposit or its distribution, except possibly at 20 lb./sq. in., the minimum pressure at which the apparatus will operate, when slightly heavier deposits were obtained. With petroleum distillate sprays, however, reduction in spraying pressure from the maximum (25 lb./sq. in.) decreased the weight of deposit and impaired the equality of distribution and accuracy of replication.

4. *Time of exposure to mist.*

The time of exposure of the test-papers to the mist was arbitrarily fixed at five minutes after spraying. It seemed from the previous experiments that, whereas the aqueous spray delivered at 25 lb./sq. in. spraying pressure and at 20°C. produced a mist which had settled completely within that time, the petroleum distillate spray under similar conditions produced a mist part of which was still in suspension in the atmosphere of the tower after the five minutes had elapsed. It was thought desirable, therefore, to measure the increase in weight of deposit obtained when the test-papers were exposed to mists of the petroleum distillate and aqueous sprays for different periods of time. A series of tests was carried out with the two dye

TABLE X.

5 cc. of 1 per cent. petroleum distillate solution of " Oil Golden Yellow 13777 " sprayed at 25 lb./sq. in. in 13·2–13·6 secs. at 20°C. Test-papers exposed for periods of from 0 to 6 mins. after spraying. Interval between sprayings 10 mins. One priming dose of 5 cc.

Time of exposure of Test-Papers in minutes after spraying	0	0·5	1	2	3	4	5	6
Mean weight of dye deposited in mg. per Test-Paper	0·038	0·139	0·199	0·248	0·291	0·316	0·324	0·338

TABLE XI.

5 cc. of 1 per cent. aqueous solution of " Orange 60506 " sprayed at 25 lb./sq. in. in 11·5–11·9 secs. at 20°C. Test-papers exposed for periods of from 0 to 6 mins. after spraying. Interval between sprayings 10 mins. Five priming doses of 5 cc. each.

Time of exposure of Test-Papers in minutes after spraying	0	0·5	1	2	3	4	5	6
Mean weight of dye deposited in mg. per Test-Paper	0·110	0·259	0·296	0·322	0·347	0·356	0·355	0·362

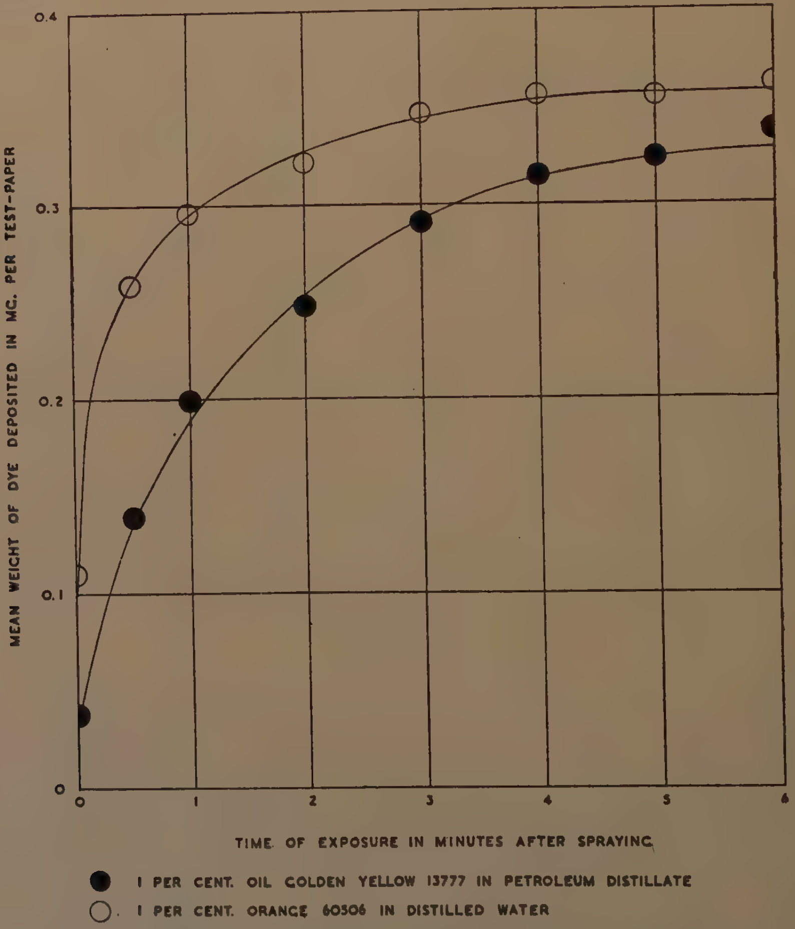

Fig. 6.—Graph showing the relationship between the weight of deposit and the time of exposure of the test-papers to the falling mist.

solutions at 25 lb./sq. in. spraying pressure and 20°C. and test-papers were exposed to the mist for periods of time from 0 minutes, *i.e.* papers removed immediately after spraying, to 6 minutes after spraying. The mean weights of deposit per test-paper obtained at the different times of exposure are given in Tables X and XI and are shown graphically in fig. 6.

It is seen that with the petroleum distillate spray, from a low initial weight obtained when the test-papers are removed immediately after spraying, the weight of deposit rises gradually as the time of exposure increases and is still rising, although slowly, after 6 minutes' exposure. With the aqueous spray, the initial deposit is heavier than that obtained with the petroleum distillate spray and increases in weight with time of exposure far more rapidly until four minutes after spraying when there is no further increase in weight. It is clear, therefore, that, with an aqueous spray, five minutes' exposure of test-papers after spraying is sufficient to ensure that a maximum deposit is obtained. With a petroleum distillate spray in this time the great majority of the droplets have settled. It seems that five minutes' exposure, although chosen arbitrarily, is a suitable time for both types of spray. Furthermore, these results show that, as surmised in the previous section, there is a margin with the aqueous spray for a reduction in the droplet size of the mist without loss in weight of deposit as the entire mist settles in four minutes and, until the droplets are so reduced that they do not settle in five minutes, no difference in weight of deposit will be observed. Mists produced from petroleum distillate sprays, however, take more than five minutes to settle and thus any alteration in droplet size will mean that more or less of the mist will be deposited in five minutes and that the weight of deposit will be sensitive to any factor tending to influence droplet size.

Unfortunately the stock of petroleum distillate used in the previous tests had become exhausted and no sample remained on which estimations of its physical characters could be made. The petroleum distillate used in this and subsequent tests evidently differed slightly from that previously used and gave weights of deposit lower than those already obtained when sprayed under similar conditions: the physical characteristics of this second sample were as follows :—

Sp. Gr.=0·786 at 20°C. Flash Point=111°F.

Distillation Range.
°C. ... ... 170, 180, 190, 200, 220, 240, 260.

Percent. vol. distilled 5, 24, 44, 56, 76, 90, 98.

5. *Volume of liquid sprayed.*

Although it is obvious that the weight of deposit obtained from any spray will depend on the volume of liquid sprayed it is not known whether there is a direct relationship between the volume of liquid and the weight of deposit. Should such a relationship exist then an alteration of the volume of liquid sprayed would be a convenient method of controlling the weight of the deposit. To determine the weights of deposit given by different volumes of liquid a series of spraying tests was carried out with the petroleum distillate and aqueous dye solutions in which 1–8 cc. was sprayed at 20°C. and 25 lb./sq. in. spraying pressure. The results of these tests are given in Tables XII and XIII and in fig. 7.

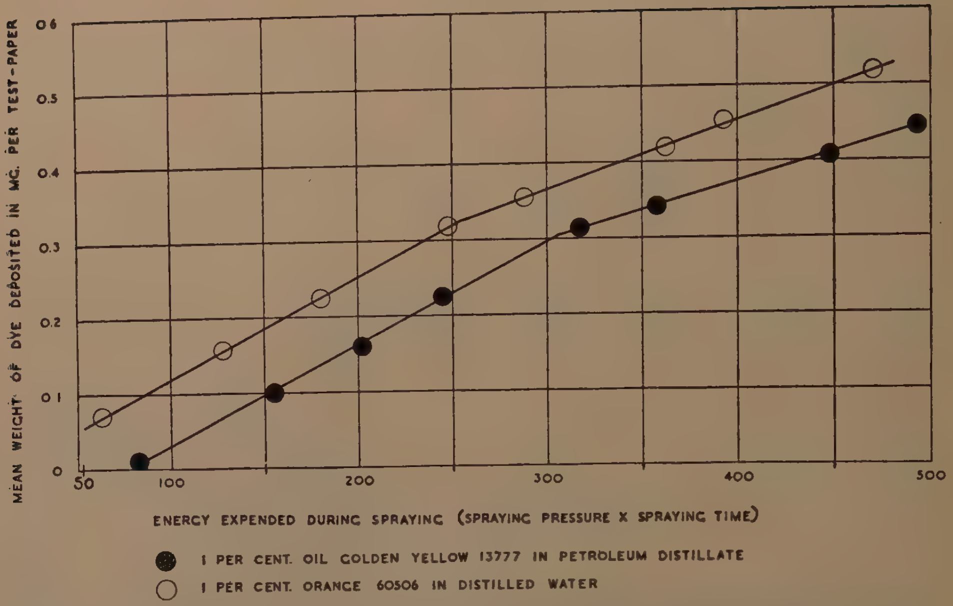

Fig. 7.—Graph showing the relationship between the weight of deposit obtained from increasing volumes of liquid and the energy expended during spraying.

TABLE XII.

1–8 cc. of 1 per cent. petroleum distillate solution of " Oil Golden Yellow 13777 " sprayed at 25 lb./sq. in. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. One priming dose of 5 cc.

Vol. of liquid sprayed in cc.	1	2	3	4	5	6	7	8
Time to spray through in seconds ...	3·3	6·2	8·1	9·8	12·7	14·3	17·9	19·7
Mean weight of dye deposited in mg. per test-paper	0·010	0·100	0·161	0·227	0·315	0·342	0·407	0·445

TABLE XIII.

1–8 cc. of 1 per cent. aqueous solution of " Orange 60506 " sprayed at 25 lb./sq. in. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. Five priming doses of 5 cc. each.

Vol. of liquid sprayed in cc. ...	1	2	3	4	5	6	7	8
Time to spray through in seconds ...	2·5	5·1	7·2	9·9	11·5	14·5	15·7	18·8
Mean weight of dye deposited in mg. per test-paper	0·072	0·156	0·223	0·318	0·353	0·419	0·454	0·518

With both the petroleum distillate and the aqueous sprays neither the weights of deposit nor the spraying times were proportional to the volume of liquid sprayed. There was, however, a direct relationship between the weights of deposit and the energy (spraying pressure × spraying time) expended during spraying (or spraying time alone as spraying pressure was constant throughout the series). In fig. 7 the weights of deposit given by 1–8 cc. of dye solution are plotted against the energy expended during spraying. The graphs obtained follow a straight line for the points representing 1–4 cc. of liquid and then abruptly change direction for the points for 5–8 cc. indicating that at a point between 4 and 5 cc. the increment in weight of deposit is decreased by a constant amount. It is suggested that this is due to a partial displacement of spray-laden air through the aperture at the top of the tower by the air stream from the atomiser. It presumably requires the spray from between 4 and 5 cc. of liquid to fill the tower and it is not until this is accomplished that spray droplets are lost by displacement.

The results obtained with the aqueous spray were essentially similar to those with petroleum distillate except that heavier deposits were obtained throughout. The graphs for values up to 4 cc. of both sprays are parallel but for values above 4 cc. they diverge slightly. It is evident, therefore, that the amount of spray lost, presumably by displacement, is greater in the case of the petroleum distillate spray than the aqueous spray, a point readily understood in the light of the greater persistence of petroleum distillate mists. The fact that the graphs for the lower values are parallel shows that the increment in weight of deposit with increasing volume sprayed is the same for both liquids. The heavier total deposit obtained with the aqueous spray, however, is due to either a smaller volume required to raise the vapour pressure within the tower to saturation point or the fact that partial evaporation of water droplets does not prevent their settling within 5 minutes after spraying to the same degree as obtains with the smaller petroleum distillate droplets. This is shown by the weights of deposit obtained when 1 cc. of each liquid is sprayed; 1 cc. of petroleum distillate dye solution gives a deposit of 0·01 mg. of dye per test-paper, whereas 1 cc. of aqueous dye solution gives a deposit of 0·072 mg.

The irregularity in the times taken to spray through the different volumes of liquids was due to the gravity feed which was used on the atomiser. An important contributory factor to the rate of spraying is the height of the "head of liquid" standing in the reservoir. Owing to the shape of the reservoir and its duct (see fig. 1), the "head of liquid" is not proportional to the volume to be sprayed. Although this type of reservoir is adequate where the volume of liquid is kept constant, where it is desirable to vary the volume the gravity feed should be replaced by a suction feed. When this improvement has been made there would seem to be no reason why the weight of deposit should not be controlled by the volume of liquid sprayed.

6. *Restriction of flow of liquid.*

The previous experiments have indicated that when a liquid is sprayed at a constant temperature the droplet size of the mist tends to be proportional to the energy expended during spraying and that, within limits which have already been defined, the weight of deposit is a function of the droplet size of the mist. Thus a restriction in the rate of flow of the liquid into the atomiser, by increasing the time taken to spray through, should result in a greater expenditure of energy during spraying and a reduction in the weight of deposit obtained. To demonstrate this, 5 cc. of petroleum distillate and aqueous dye solutions were sprayed at 20°C. and 25 lb./sq. in. spraying pressure at nine consecutive positions of the needle-valve controlling the rate of delivery of the liquid from fully open to almost closed. Tables XIV and XV give the times taken to spray the liquids at each position of the needle valve and the mean weights of dye deposited per test-paper. These results are illustrated graphically in fig. 8.

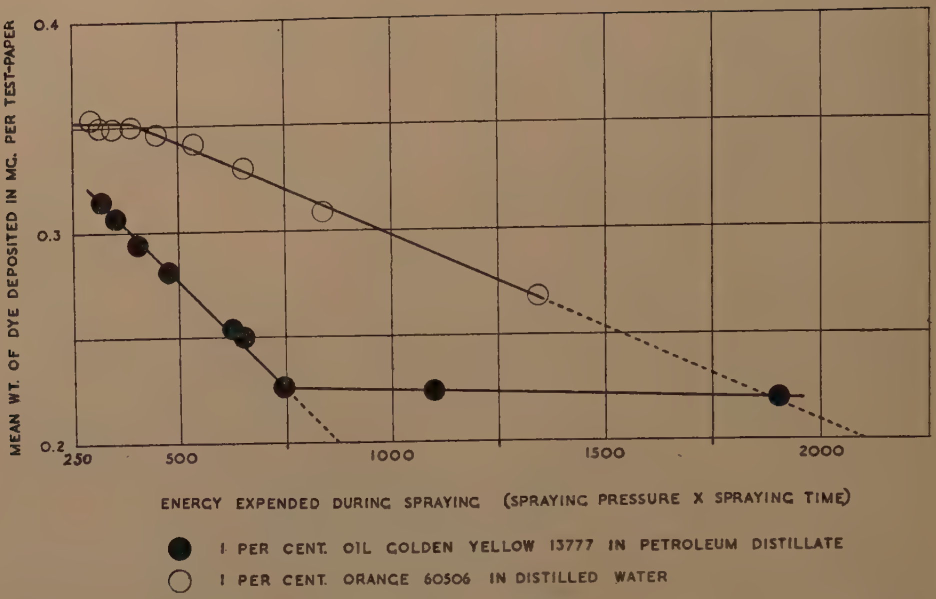

Fig. 8.—Graph showing the relationship between weight of deposit and spraying time when spraying pressure and volume of liquid are constant.

TABLE XIV.

5 cc. of 1 per cent. petroleum distillate solution of "Oil Golden Yellow 13777" sprayed at 25 lb./sq. in at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. One priming dose of 5 cc.

Time to spray through in secs.	12·7	14·0	16·1	19·0	25·5	26·3	29·8	44·0	76·3
Mean weight of dye deposited in mg. per test-paper ...	0·315	0·307	0·294	0·282	0·254	0·250	0·226	0·224	0·219

TABLE XV.

5 cc. of 1 per cent. aqueous solution of "Orange 60506" sprayed at 25 lb./sq. in. at 20°C. Test-papers exposed for 5 mins. after spraying. Interval between sprayings 10 mins. Five priming doses of 5 cc. each.

Time to spray through in secs.	11·7	12·5	13·7	15·5	18·0	21·5	26·2	33·5	53·7
Mean weight of dye deposited in mg. per test-paper ...	0·358	0·350	0·349	0·350	0·346	0·342	0·330	0·309	0·268

It was found that with spraying times up to 30 seconds the weight of deposit from the petroleum distillate dye solution decreased rapidly, varying inversely with the spraying time or energy expended (see Table XIV and fig. 8). At spraying times from 30 seconds to over 1 minute, however, only a slight reduction in weight of deposit was obtained. With the aqueous dye solution at spraying times up to 16 seconds the weight of deposit remained constant, but it declined, though more slowly than the deposits from petroleum distillate, at spraying times above 16 seconds, the weight of deposit again varying inversely with the spraying time or energy expended (see Table XV and fig. 8). At the maximum spraying time possible with the apparatus, *i.e.* when the needle-valve was almost closed, the weight of deposit, unlike that from the petroleum distillate spray, was still falling in relation to the energy expended. These results may be interpreted in the light of the conclusions drawn from the earlier experiments. The effect of increasing the spraying time is to increase the amount of energy available for atomising the 5 cc. of liquid and thus to break the liquid into finer and finer droplets. The smaller the size of the droplets the longer they take to settle in the tower and, if they do not settle completely within five minutes of spraying, the greater the number still left in suspension when the test-papers are removed and the smaller the weight of deposit collected on the test-papers. It has been shown, with petroleum distillate dye solution sprayed at 20°C. and 25 lb./sq. in. with the needle-valve fully open, that not all the droplets settle within 5 minutes of spraying ; thus the reduction in droplet size due to increased spraying time causes an immediate fall in weight of deposit. With the aqueous dye solution sprayed at 20°C. and 25 lb./sq. in. with the valve open the droplets settle completely in four minutes and can be reduced in size until they settle in five minutes before a corresponding fall in weight of deposit ensues ; hence there is no reduction in weight of deposit from the aqueous spray until spraying time is increased above 16 seconds. The relatively slow decrease in weight of deposit from the aqueous spray at spraying times above 16 seconds, as compared with the rapid decrease obtained with the petroleum distillate spray, presumably indicates that the energy expended in producing droplets of a given size was greater in the case of the former. The ratio between the quantities of energy involved in both instances may be estimated by continuing the graphs (see fig. 8, broken line) representing the fall in weight of deposit with energy expended for the two sprays until they cut the abscissa. The ratio between the distances of these two points of intersection from the origin is 0·42 and represents the ratio between the quantities of energy required to atomise the petroleum distillate and the aqueous dye solutions to the same degree. This ratio is evidently determined by the surface tension of the liquids. The surface tension at 20°C. of the petroleum distillate dye solution was between 25 and 25·5 dynes/cm. while that of the aqueous dye solution was 60 dynes/cm. giving a ratio of 0·42. The sharp change in direction of the line of the graph for the petroleum distillate spray at 30 seconds spraying time (see fig. 8) would seem to indicate that a point is reached when an increase in the amount of energy available for atomisation produces no further reduction in droplet size under these conditions of spraying : the slight decrease in weight of deposit at spraying times in excess of 30 seconds is attributable to displacement of mist from the tower by the air-stream from the atomiser. This limitation of droplet size, too, may be a function of the surface tension of the liquid. It is of interest that a point where the water spray behaved in a similar manner was not reached ; thus it is not known whether the limit for size of water droplets, if it exists, coincides with that of the petroleum distillate droplets.

Thus, over a certain range, the droplet size of the mist governs the weight of deposit obtained with both petroleum distillate and aqueous dye solutions. Outside this range, *i.e.* where droplets are either too large to remain in suspension for five minutes or have reached a limit of subdivision, the weight of deposit is unaffected by factors tending to produce small variations in droplet size. It seems desirable, therefore, where close replication of deposits is essential, to adjust the spraying time and pressure to produce mists with droplets ranging within one or the other of these limits of size. In the foregoing experiments the aqueous dye solution sprayed at 25 lb./sq. in. and delivering 5 cc. of liquid in under 16 seconds produced a mist within the upper range of droplet size and thus the deposits obtained were insensitive to small changes in spraying pressure, spraying time, temperature or time of exposure of the test-papers to the mist. The petroleum distillate dye solution sprayed under similar conditions produced a mist within the range of droplet size governing weight of deposit and thus the deposits were sensitive to any factor tending to alter droplet size. This disadvantage can be eliminated by increasing the spraying time until the droplet size is at a minimum when small variations in pressure and spraying time will leave both the droplet size and the weight of deposit unaffected. With any liquid to be sprayed, given its surface tension, it should be possible to decide whether it is preferable to spray coarse or fine droplets to obtain the most efficient replication of deposits.

Construction of the Testing Chamber.

The spraying tower which has been described was designed to coat panels 20 cm. square with an even deposit of insecticidal material of known weight. These panels

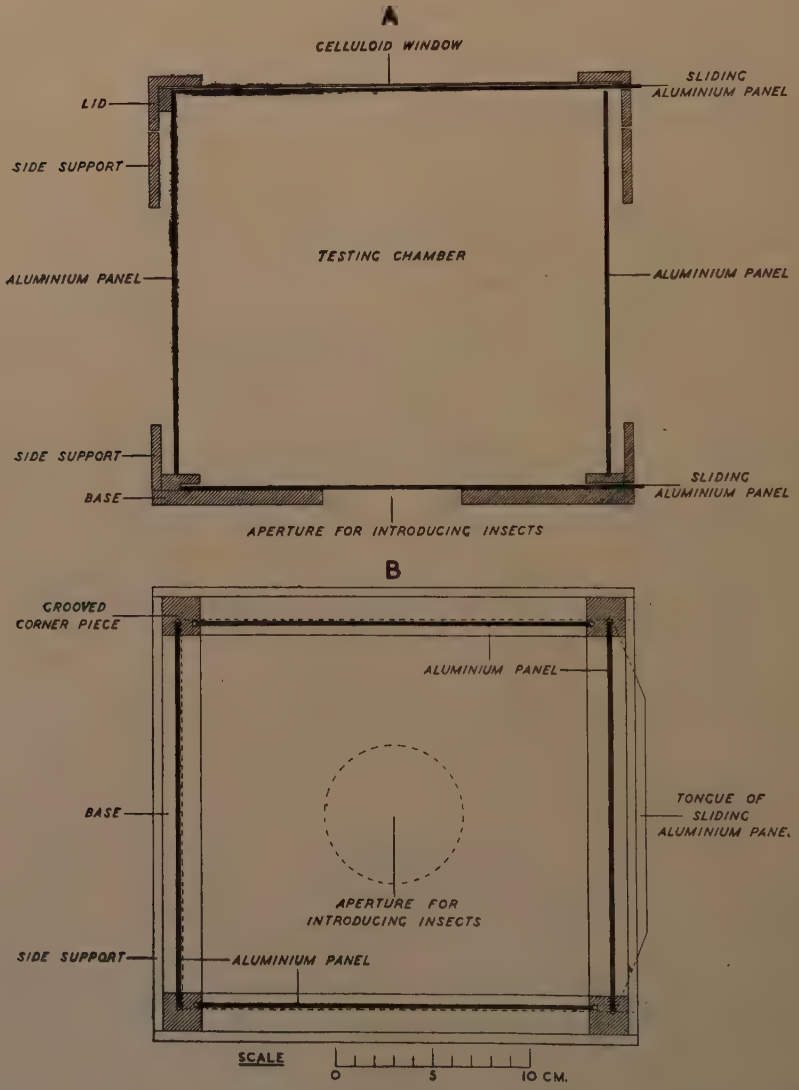
A
CELLULOID WINDOW
LID
SLIDING ALUMINIUM PANEL
SIDE SUPPORT
TESTING CHAMBER
ALUMINIUM PANEL
ALUMINIUM PANEL
SIDE SUPPORT
BASE
SLIDING ALUMINIUM PANEL
APERTURE FOR INTRODUCING INSECTS
B
GROOVED CORNER PIECE
ALUMINIUM PANEL
BASE
TONGUE OF SLIDING ALUMINIUM PANEL
APERTURE FOR INTRODUCING INSECTS
SIDE SUPPORT
ALUMINIUM PANEL
SCALE
0
5
10 CM.

Fig. 9.—A. The testing chamber seen in longitudinal section ; B. A plan of the testing chamber with the lid removed.

can then be built up into a cubical testing chamber into which insects such as house-flies, mosquitos, cockroaches or bedbugs can be introduced and observed at stated intervals. The panels used with this apparatus were made of aluminium sheet $\frac{1}{16}$in. thick. Each set of six panels forming the sides of the chamber comprises four 20 cm. square and two 20 cm. × 23·5 cm. with two corners cut away (see fig. 9, B) to form a tongue. The panels slide into grooves in a wooden crate the construction of which is shown in fig. 9 and Pl. I, figs. 2 & 3. The base of the crate is a square piece of board in the centre of which is a circular hole through which the insects are introduced into the chamber. Arising from the corners of the base are four pieces of wood 20 cm. high, square in section and bearing grooves on their two inner faces into which slide the square panels. These panels form the walls of the chamber. The corner pieces are capped by a lid, in the top of which is a sheet of celluloid. One of the tongued panels slides in grooves immediately above the base and forms the floor of the chamber, while the other passes through a slot in the lid and, resting upon the tops of the vertical panels, forms the ceiling of the chamber. When in position, the outer surface of the upper sliding panel is in contact with the celluloid window of the lid. The lower edge of the slot in the lid is slightly curved downwards and the vertical panel beneath this slot, by resting in a shallow groove in the base, is set slightly lower than its fellows to give clearance for the sliding panel so that the insecticidal deposit on its surface is not disturbed when the panel is pushed in or drawn out. The tongues project from the side of the crate and provide a finger hold for withdrawing the panels. In practice the four vertical panels are dropped into their grooves and the lid placed in position. The insects are introduced into the chamber through the hole in the base, counted approximately and the aperture closed by inserting the lower tongued panel. The upper tongued panel is then inserted into the lid. At suitable intervals the upper tongued panel is partially withdrawn and the condition of the insects observed through the celluloid window. Observations are continued until all the insects are unable to fly or are otherwise immobilised. The aluminium panels may be covered with any surface such as wall paper, paint or plaster board, except that where the material occupies an appreciable thickness, as plaster board does, this can only be mounted upon the four vertical panels, the tongued panels being left untreated. Sprayed panels may be stored under different conditions of weathering and the efficiency of the insecticidal deposits determined after various periods of storage.

Summary.

The construction of a spraying tower for obtaining even deposits of insecticide of known weight is given, and the method of adjustment of this apparatus and procedure adopted during spraying are described.

To determine the weight of deposit obtained with different liquids under various conditions of spraying, a colorimetric method of estimation was adopted. Deposits of dye solution were collected on test-papers disposed at different positions on the area sprayed, the dye dissolved in 50 cc. of solvent and the concentration of dye in this solution measured by means of the Spekker Photo-electric Absorptiometer. By this method estimations of the weight of dye deposited on each test-paper were made to 0·01 mg. with great rapidity. The solutions of dye used in these experiments were 1 per cent. wt./vol. solution of " Oil Golden Yellow 13777 " in petroleum distillate and a 1 per cent. wt./vol. aqueous solution of " Orange 60506 ".

Series of tests were carried out with both the petroleum distillate and the aqueous dye solutions to determine the replication and distribution of deposits obtained with the spraying tower. It was found that the Standard Deviation of the mean weight of deposit per test-paper for the two spray liquids, both from one test to another and between different positions on the area sprayed in any one test, fell between ±0·8 per cent. and ±3·0 per cent. of the mean under the most favourable conditions for spraying.

The effect of varying the factors governing weight of deposit was studied and from the results certain general conclusions were drawn. Firstly, where the surface to be sprayed is removed from the tower before the mist has completely settled and the time of exposure of the surface to the mist is constant, the weight of deposit varies directly with the degree of atomisation of the liquid sprayed ; the smaller the droplet size of the mist the lighter the deposit obtained. Secondly, the droplet size of the mist varies directly with the surface tension of the liquid sprayed and inversely with the energy (the spraying pressure × the spraying time) expended in atomising a given volume of liquid. Thus any factor tending to alter the surface tension of the liquid, the spraying pressure or the spraying time will alter the droplet size of the mist and affect the weight of deposit obtained within the limits defined. Where the mist has settled completely before the sprayed surface is removed from the tower, small variations in droplet size of the mist appear to have no effect on the weight of deposit. With the petroleum distillate spray the amount of energy expended in atomising 5 cc. of the liquid was progressively increased until a point was reached when the weight of deposit remained almost constant even when the amount of energy available was more than doubled. It is suggested that at this point no further reduction of droplet size is possible under the conditions of spraying. Small variations in either spraying time or pressure should have no effect on the weight of deposit obtained where droplet size of the mist is at a minimum.

An important factor governing the rate of evaporation of the spray droplets and hence the weight of deposit is the degree of saturation of the atmosphere within the tower with vapour of the liquid sprayed. This factor was controlled by spraying a sufficient number of priming doses of liquid to bring the vapour pressure within the tower to near saturation point and by adopting a standard interspraying time so that the amount of vapour lost to the outside atmosphere between consecutive sprayings was always the same. In this way it was assured that the same partial pressure of vapour of the liquid to be sprayed obtained within the tower at the commencement of each spraying. At 20°C., one priming dose was found to be necessary for the petroleum distillate spray and five for the aqueous spray.

The effect on the weight of deposit of a rise in temperature from 20° to 30°C. differed with the petroleum distillate and the aqueous sprays. In the case of the former, the weight of deposit obtained at 30°C. was less than that at 20°C., but with the aqueous spray there was no significant difference at either temperature. A rise in temperature by decreasing the surface tension and viscosity of the liquids sprayed would be expected to cause a reduction in droplet size of the mist. It is suggested that this may be proportionately greater in the case of the petroleum distillate spray than the aqueous spray. Even if this is not so, as the mist from the aqueous dye solution sprayed at 20°C., 25 lb./sq. in. spraying pressure and minimum spraying time, unlike that from the petroleum distillate dye solution, settled within the five minutes for which test-papers were exposed, further reduction in size of the aqueous spray droplets can take place before some of them remain suspended for longer than five minutes and cause a fall in weight of deposit. Thus it would be expected that with a rise in temperature there would be a greater reduction in weight of the deposit from petroleum distillate than from aqueous sprays. It is, therefore, important to control temperature where petroleum distillate sprays are used.

A reduction in the spraying pressure from 25 lb./sq. in. to 22·5 and 20 lb./sq. in. increased the minimum spraying time for 5 cc. of liquid to a different degree for both the petroleum distillate and the aqueous sprays. It was found with the former spray that the weight of deposit varied inversely with the energy (spraying pressure × spraying time) expended in atomisation, and that the energy used increased and the weight of deposit fell with decreasing spraying pressure. With the aqueous spray the increase in energy with decreasing spraying pressure was very much less and no fall in weight of deposit was observed : this is evidently due to the fact that at 20 lb./sq. in. the aqueous droplets still settled within five minutes after spraying. Thus, for accurate replication of deposits from petroleum distillate sprays, the spraying pressure should be rigidly controlled, but for aqueous sprays this is not so important.

By exposing test-papers to mists for increasing periods of time after spraying, it was shown that whereas at 20°C., 25 lb./sq. in. and minimum spraying time the mist produced from the aqueous dye solution had settled completely in four minutes, the mist from the petroleum distillate solution was still partially in suspension six minutes after spraying. Thus with a standard time of exposure to the mist of five minutes for both types of spray there is a margin for reduction in droplet size of the aqueous mist without loss in weight of deposit which is not present in the petroleum distillate mist. The weight of deposit from a petroleum distillate mist, therefore, will be sensitive to any factor tending to influence droplet size.

When volumes of liquid from 1 to 8 cc. were sprayed at 20°C., 25 lb./sq. in. and minimum spraying times it was found that the weights of deposit were proportional to the energy (spraying pressure x spraying time) expended during spraying, but that neither the weight of deposit nor the spraying time was proportional to the volume of liquid sprayed. Furthermore, when a volume of liquid greater than 5 cc. is sprayed the increase in deposit with spraying time falls by a constant amount. This is presumed to be due to partial displacement of spray-laden air by the air stream from the atomiser which takes place after 4–5 cc., the amount required to fill the tower with mist, has been sprayed. The disparity between volume of liquid on the one hand and the spraying time and weight of the deposit on the other was due to the gravity feed of the atomiser in which the " head of liquid " standing in the reservoir was not proportional to the volume. This difficulty could be overcome by introducing a suction feed. Throughout these series a heavier deposit was obtained from the aqueous than from the petroleum distillate spray. This may have been due to a difference in the amount of spray evaporated. It is more probable, however, that given the same amount of evaporation from droplets of both liquids, a higher proportion of the water droplets still remained sufficiently heavy to settle and deposit the non-volatile dye within five minutes of spraying than the petroleum distillate droplets which initially are relatively small.

A restriction of the rate of flow of liquid through the atomiser by closing the needle-valve and reducing the effective aperture of the fluid jet, prolonged the time taken to spray a given volume of liquid and thus increased the amount of energy available for atomisation. This resulted in a reduction in the droplet size of the mist and produced a fall in the weight of deposit. With the petroleum distillate spray, the weight of deposit fell in inverse proportion to the energy expended until a spraying time of about 30 seconds was reached and then remained practically constant although the energy available was more than doubled. It seems, therefore, that there is a limiting droplet size under the conditions of the spraying and that this was reached at 30 seconds spraying time. With the aqueous spray the weight of deposit remained constant as the energy expended increased until the droplets became sufficiently small for some of them to remain in suspension after the test-papers were withdrawn from the tower, whereupon the weight of deposit began to fall, though more slowly than that from the petroleum distillate spray, in inverse proportion to the quantity of energy used. No limiting value for droplet size was reached for the aqueous spray. The rates of fall in weight of the deposits from the two sprays, *i.e.* the energy needed in each case to produce droplets of given size, were found to be in the same ratio as the surface tensions of the two liquids.

It was concluded that with petroleum distillate sprays delivered at 20°C., 25 lb./sq. in. spraying pressure and minimum spraying time it is necessary to exercise close control over these variables and over all details of spraying procedure if accuracy of replication of deposits is to be achieved. It may be possible, however, to relax these precautions if the sprays are delivered at spraying times above 30 seconds, *i.e.* with the needle-valve partially closed, when droplet size of the mist is at a minimum. With aqueous sprays delivered at 20°C., 25 lb./sq. in. spraying pressure and minimum spraying time, small variations in spraying conditions do not seem to affect the accuracy of replication or equality of distribution of the deposits.

The spraying tower described was designed to coat panels 20 cm. square with an even deposit of insecticidal material of known weight. These panels can then be built up into a cubical chamber for testing residual deposits of insecticides against such insects as houseflies, mosquitos, cockroaches or bed-bugs. The construction of this chamber and its method of use are described.

Acknowledgements.

[Musca domestica, L.]

The writer wishes to express his thanks to Mr. D. A. Lambie for his advice and criticism and for carrying out all determinations with the Spekker Photo-electric Absorptiometer ; also to Mr. D. J. Finney who gave advice on the statistical treatment of the results and to Mr. D. S. Kettle who kindly checked the calculations and read the manuscript.

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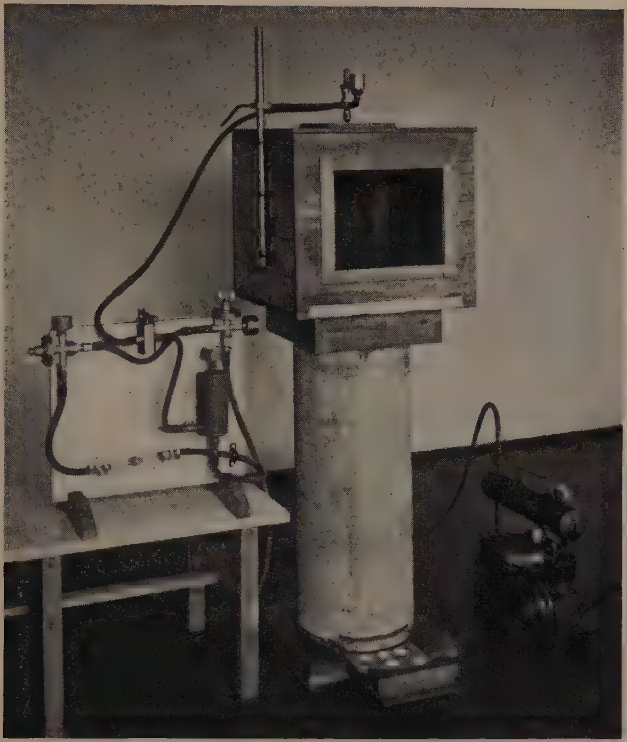

FIG. 1. The Spraying Tower showing the sliding tray bearing the test plate partially withdrawn. The apparatus for controlling the air stream is seen on the left and the ¼-h.p. compressor on the right of the tower.

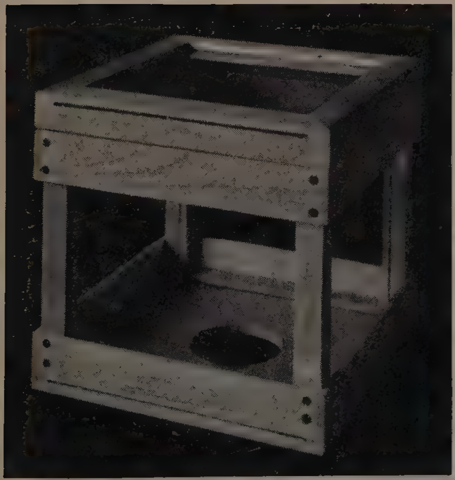

FIG. 2. The testing chamber with the aluminium panels removed.

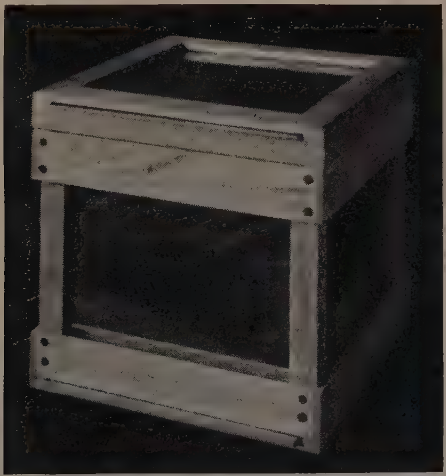

FIG. 3. The testing chamber with the aluminium panels in position.

SOME EAST AFRICAN SPECIES OF *LYGUS*, WITH NOTES ON THEIR HOST PLANTS.

By T. H. C. Taylor, D.Sc.

Imperial Institute of Entomology.

In my recent paper on " *Lygus simonyi*, Reut., as a Cotton Pest in Uganda "*, I referred in the introductory notes to the fact that I had studied the male genitalia and other characters of about 20 species of *Lygus* in order to determine reliably all specimens collected in the course of the field work on which I was then engaged. In the present paper all of these species are described. Only a few of them occur commonly on cotton, and only one, which must now be called *L. vosseleri*, Popp., is a major pest of cotton, but the others were encountered, in most cases very frequently, in connection with an investigation of the alternative host plants of the cotton-frequenting species, and two are common on coffee (with *L. coffeae*, China, which belongs to a different group of species and is therefore not included here). Some of them bear a very close resemblance to *L. vosseleri* and often occur with it on the same plants and in the same localities ; consequently considerable confusion had existed previously as to the identity of the various species collected.

In connection with field work it is often necessary to identify large numbers of specimens rapidly and without adequate facilities for the examination of genitalia. Consequently, the object of my study of the external genitalia of the males was not only to separate the species on the basis of these structures but also to test by means of them the reliability for diagnostic purposes of the few differences, such as those of colour and markings, that occur in both sexes and are readily visible with a hand lens. I concluded that the male genitalia afford the best characters for determination whenever they can be examined, but that colour and markings can suffice in both sexes for this purpose, despite the great variation which occurs in most species. The only other characters that are of value for the separation of species, within the group with which I have dealt, are size and (occasionally) length of rostrum. There is, in fact, a remarkable uniformity throughout this species group, so that in the absence of males or of facilities for examining genitalia, determination must depend upon a combination of a few simple characters, two of which, colour and markings, are variable, often difficult to describe, and can be utilised satisfactorily only after experience has been gained of many species and specimens from numerous localities and plants.

The species described in this paper constitute what may be called the *simonyi* group of *Lygus* and have the following features in common :—

1. A smooth, semi-shiny appearance (never very shiny because of slight pubescence and minute punctation).

2. The whole of the dorsal surface, including the hemelytra, and also the ventral surface, rather sparsely covered with very fine, short decumbent hairs directed backwards.

3. The whole of the dorsal surface, including the hemelytra, indistinctly and minutely punctate ; the scutellum and (to a lesser extent) the pronotum very slightly rugose transversely.

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*Bull. ent. Res., **36**, p. 121, 1945. In a later paper (Bull. ent. Res., **37**, p. 503, 1947) I pointed out that the correct name of this insect is *L. vosseleri*, Popp., not *L. simonyi*, Reut., the latter being a species which does not breed on cotton and is rarely found on it.

4. The head, in dorsal view, short, very markedly transverse and very closely apposed to the pronotum.

5. The left paramere of the male with a toothed, laminate process (except in two species) and a relatively long and slender stem.

Other African species, in addition to the well-known *L. coffeae*, of which I have examined type or reliably named specimens from the British, Paris or Helsinki Museums, and which also do not belong to this group, are *incertus*, Popp., *conspersus*, Wlk., *pallidulus*, Wlk., *illepidus*, Wlk., *suffusus*, Wlk., *neavei*, Popp., *sjöstedti*, Popp., *suturellus*, Popp., *flaviventris*, Popp., *nigriscutum*, Popp., *alpicola*, Popp., *gabonius* (Kirk.), *nigropunctatus*, Popp., and *meruensis*, Popp. Some of these species have one or two of the features characteristic of the *simonyi* group, but none has all of them ; moreover, their general form, which is an important feature, is different. Those of which I have been able to examine males have the left paramere relatively short and stout and lack the laminate process, which, above all the other characters, I regard on present evidence as a criterion of the *simonyi* group whenever it is present, though its absence does not necessarily exclude a species from this group.

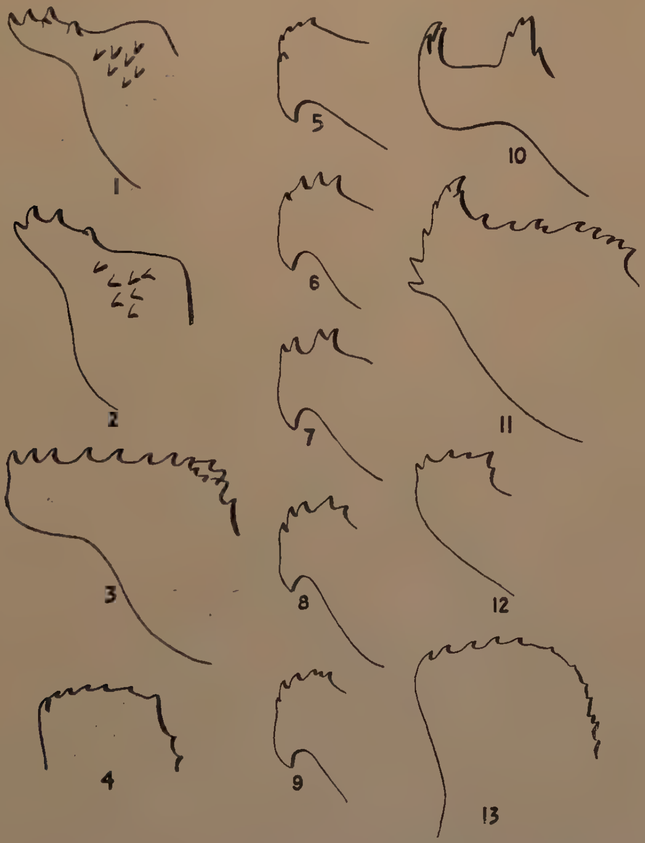
L. thomasi, Reut., which I have not collected myself, certainly belongs to this group, and I have therefore included a redescription of it in this paper, based on material from the Helsinki and Paris Museums.

The species with which I have been concerned, with only two exceptions (*thomasi*, Reut., and *indecorus*, sp. n.), were all collected in Uganda, and my descriptions of them are based on Uganda series only. Most or all of them doubtless occur in neighbouring territories and a few of them are known from many parts of Africa, from the Sudan to South Africa. The variation in colour and in distinctness and extent of markings occurs not only within one locality but also between series from different territories and types of country, and due allowance must be made for this in interpreting my key and descriptions which cover the range of variation within Uganda but not necessarily within the African continent. The genus is very inadequately known from Africa as a whole, few collectors having been attracted by it, and the material available from African countries other than Uganda is not yet sufficient to enable me to undertake a more extensive survey.

I have described only those species of which I obtained series sufficiently long for the study and dissection of genitalia. There are, in various collections, other new African species which are represented by single or by only a few specimens, and these I have preferred to leave undescribed for the present. To describe a species of a genus such as *Lygus* from scanty material is rash and often harmful, except in those cases which show strikingly unusual characters, but unfortunately many descriptions of species by certain earlier workers were based on single or a few mutilated specimens, and their descriptions are unreliable in consequence. Further, most of these workers described their species in great detail without stressing the few features that are of specific value, and since most of the details given apply to the whole group it is impossible to recognise with certainty many of the species from the descriptions. In these circumstances, I have thought it desirable to redescribe those few species in my own collection which have already been described from material from other sources by other authors, and of which I have seen type or reliably named specimens. The remaining species are described as new, and I have little doubt that they are actually new, since they are not identical with any named specimens which I have been able to examine, and disagree, particularly as regards size, with the descriptions of those African species of which I have not been able to see named specimens. Nevertheless, the possibility that one or two of them may prove later to be synonymous with species of other authors must not be overlooked.

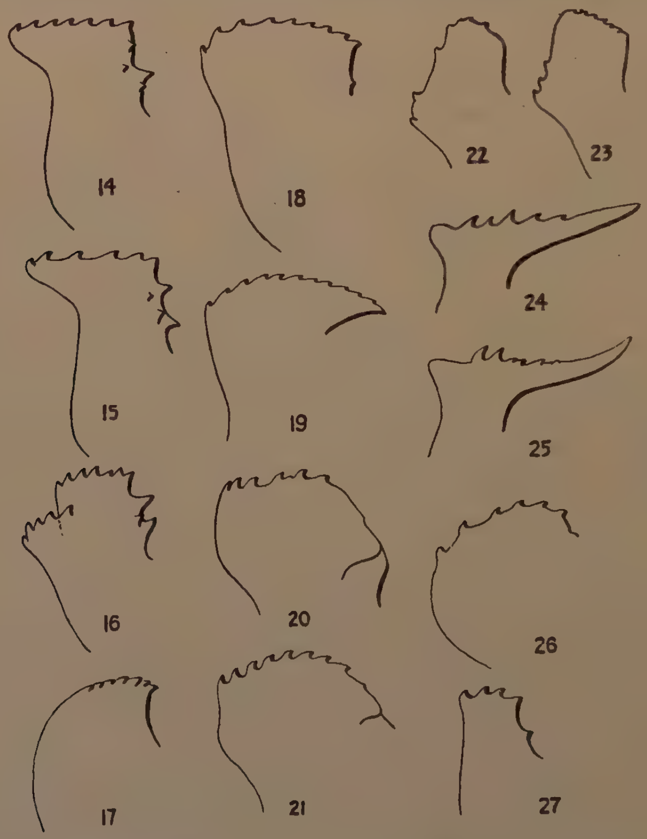
In the male genitalia of this group of *Lygus*, the left paramere is exceptionally large and elaborate. Its general form is constant throughout the group (figs. 94 and

95), but in detail it varies greatly among the species. For each species its form and structure are constant and highly diagnostic. It consists of a swollen basal portion, which bears a dentate, laminate process and from which arises a relatively long, curved stem terminating in a hook. The stem and the basal portion always bear strong setae and sometimes teeth. The precise shape of the paramere as a whole, the distribution and arrangement of the setae, and of the teeth when present, and the outline of the laminate process and the hook are all absolutely specific. Since the various parts of the paramere project in three dimensions it does not lie flat on a slide for microscopic examination, and two specimens which are being compared seldom lie in corresponding positions; consequently comparison is difficult if the whole paramere is used, and I have always preferred to cut off the hook and the laminate process, and to mount them separately on slides, as a matter of routine. These two parts suffice by themselves for the certain determination of species, and the laminate process is of special value in that it invariably projects from the body in such a way that it can readily be seen on pinned specimens without dissection, so that determination of males is always a very simple matter.



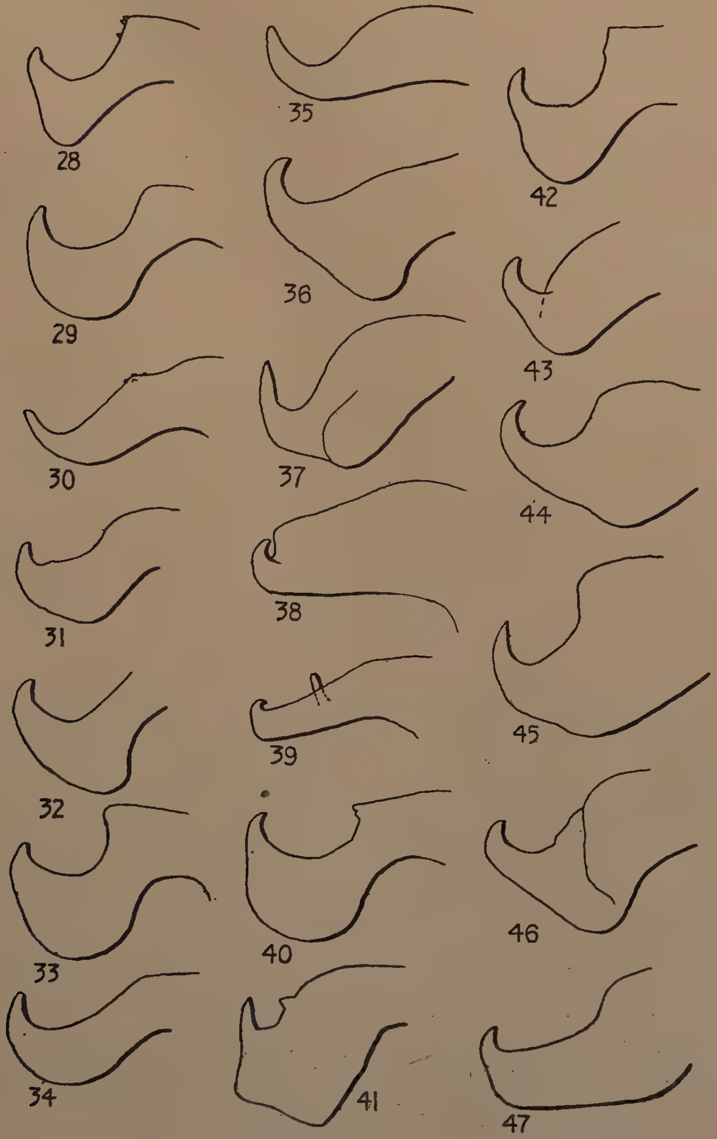
Figs. 1–13.—Laminate process of left paramere of *Lygus* spp., mounted under slight pressure.—(1 and 2) *L. ricini*, sp.n., × 150; (3) *L. vosseleri*, Popp., × 150; (4) *L. compositus*, sp.n., × 200; (5–9) *L. virens*, sp.n., ×200; (10) *L. apicalis*, Fieb., × 175; (11) *L. cupressus*, sp.n., × 165; (12) *L. entadae*, sp.n., × 175; (13) *L. nairobiensis*, Popp., × 125.

My conclusions concerning the significance from this point of view of the laminate process are based on the examination in Uganda of about 4,000 specimens. Microscope slides of the male genitalia, antennae and hemelytra of all the species here described are in the collection of the Department of Agriculture, Uganda. The drawings illustrating this paper are based on these slides. In interpreting the drawings of the laminate process, it must be borne in mind that it is actually curved and that the drawings were prepared from specimens of it that were flattened on slides. The curvature is slight, however, and does not in the least hinder the comparison of the process, in its normal position on dried specimens of the whole insect, with the drawing of it. Similarly, the illustrations of the hook are also from flattened specimens, but the flattening, while expanding and altering the outline slightly, does not make comparison any less simple. There is slight variation in the laminate process in all species, principally as regards the number of teeth, but this is never sufficient to cause any difficulty in determination. The extent of the variation is shown for seven species in figs. 1 and 2, 5–9, 14 and 15, 20 and 21, 22 and 23, 24 and 25 and 88 and 89. Although the laminate process is highly characteristic of the *simonyi* group, I have included in this group two new species, *palus* and *ugandanus*, in which the process is wanting, since they agree with my conception of the group in other respects.



Figs. 14–27.—Laminate process of left paramere of *Lygus* spp., mounted under slight pressure.—(14 and 15) *L. simonyi*, Reut., × 150; (16) *L. notatus*, sp.n., × 150; (17) *L. complexus*, sp.n., × 150; (18) *L. tenebrosus*, sp.n., × 150; (19) *L. ghesquierei*, Schout., × 120; (20 and 21) *L. arboreus*, sp.n., × 120; (22 and 23) *L. infirmus*, sp.n., × 90; (24 and 25) *L. definitus*, sp.n., × 175; (26) *L. vernoniae*, sp.n., × 150; (27) *L. chagweensis*, Popp., × 200.

The markings on the hemelytron (fig. 48 *et seq.*) are next in importance to the male genitalia for determination purposes, in spite of a general similarity almost throughout the group and the variation which occurs in most species. The drawings were mainly prepared from hemelytra cleared and mounted on microscope slides,



Figs. 28–47.—Hooked process of left paramere of *Lygus* spp., mounted under slight pressure.—(28) *L. ricini*, sp.n. ; (29) *L. vosseleri*, Popp. ; (30) *L. virens*, sp.n. ; (31) *L. apicalis*, Fieb. ; (32) *L. cupressus*, sp.n. ; (33) *L. simonyi*, Reut. ; (34) *L. notatus*, sp.n. ; (35) *L. ugandanus*, sp.n. ; (36) *L. infirmus*, sp.n. ; (37) *L. nairobiensis*, Popp. ; (38) *L. complexus*, sp.n. ; (39) *L. palus*, sp.n. ; (40) *L. tenebrosus*, sp.n. ; (41) *L. arboreus*, sp.n. ; (42) *L. definitus*, sp.n. ; (43) *L. entadae*, sp.n. ; (44) *L. compositus*, sp.n. ; (45) *L. vernoniae*, sp.n. ; (46) *L. chagweensis*, Popp. ; (47) *L. ghesquierei*, Schout.

but the markings, though often very vague, can be readily seen if the hemelytra are lifted slightly from the body with a needle so that light can shine through them. The variation is chiefly in extent and depth of colour, not in form. The important markings are those on the more strongly sclerotised parts of the hemelytron, particularly the clavus, corium and embolium, though in a few species these parts are without markings. The cuneus is useful only in that the minute, but conspicuous, apical black spot is not present in all species. The membrane is usually marked with dark infuscations enclosing hyaline patches, and most authors have described its markings, but I have found them useless as specific characters since they are almost uniform throughout the group.

The pronotum and scutellum are marked or darkly coloured in certain species, and in some there are extensive dark or black areas on the ventral surface, sometimes in both sexes and sometimes only in the males. Since these ventral markings are difficult to describe briefly, I have supplemented the descriptions with diagrams in some cases (figs. 83–87). These dark areas are particularly useful as additional features for the recognition of the economically important *L. vosseleri*.

The general ground colour is liable to considerable variation in all species. A green species may be yellowish-green or brownish-green. The antennae and legs are the same colour as the head and body, but the second antennal segment and the femora are usually dark or black distally. The length of the dark portion of the second antennal segment relative to the total length of the segment, though slightly variable in each species, is a valuable specific feature (fig. 96). The term " dark " appears frequently in my descriptions, being used to indicate a darkening, variable in actual colour but approaching black, of the general ground colour. " Dusky " is similarly used for slight darkening. All my references to colour are based on dried specimens, the colour of fresh specimens often being different and always more vivid. Teneral specimens are common in collections and are liable to be misleading ; their colour is often abnormal and markings which are normally strongly developed may be almost absent in them.

The number of specimens cited after the description of each species refers merely to the relatively short series from which the description was made. The preliminary study of the various species involved the examination of many dozens (in several cases, hundreds) of specimens of each. The dimensions given are the means of those of three specimens selected as being neither abnormally small nor abnormally large. The word " genitalia " in each description is used for brevity to refer to the drawings of the laminate process and hook of the left paramere ; the genitalia are not completely described or figured.

The types and series on which the descriptions were based, including the plesiotypes of those species of other authors which are here redescribed, have been deposited in the British Museum (except in the case of *L. thomasi*). Reliably determined duplicate series of all Uganda species are in the collections of the Department of Agriculture, Uganda.

In the notes on the plants frequented by the various species, I have adopted the term " host plant " for those plants on which breeding occurs. The adults of most, if not all, species feed on, or at least derive moisture from, a great variety of plants on which breeding does not occur, and I refer to these others as " food plants ".

I am indebted to Mr. W. E. China, of the British Museum, for much help and advice, to Mr. H. Hargreaves, formerly of Uganda, for the loan of specimens from his own collection, to Mr. H. S. Darling for sending me specimens from the Uganda collection and to the authorities of the British, Helsinki, Paris, and Vienna Museums for lending me all their African specimens of *Lygus* for study.

KEY TO THE UGANDA SPECIES OF THE *SIMONYI* GROUP OF *LYGUS*.

The following key is only tentative, since there are no doubt several species which are at present unknown, even within Uganda. It is also not entirely satisfactory because certain of the few characters available are variable and rather indefinite. It is, however, reliable within the range of species and variation known to me in Uganda, and after some consideration I have decided to publish it as a basis for future work. Since the laminate process of the left paramere affords the best means of determining the males of nearly all species, the principal purpose of the key, in which the process is rarely referred to and then only secondarily, is the determination of females. Nevertheless, it is equally applicable to both sexes.

1. Hemelytron with strong markings* in the form of a loop near costa (fig. 66) ; laminate process of left paramere wanting......................*ugandanus*, **sp.n.**

   Hemelytron with markings of different form, or without markings ; laminate process present (except in one species)..2

2. Rostrum reaching only to middle coxae ; hemelytron markings consisting at most of a faint band along claval margin of corium (fig. 80)....................*indistinctus*, **sp.n.**

   Rostrum reaching to or slightly beyond hind coxae ; hemelytron markings of different form, usually much more extensive, rarely almost wanting............3

3. Cuneus with black or dark spot at apex..6

   Cuneus without such spot, though sometimes slightly dusky at apex............4

4. Scutellum with markings, consisting of a pair of longitudinal dark lines, but these sometimes faint ; hemelytron markings consisting principally of a prominent wedge-shaped median streak (fig. 77)...............*vernoniae*, **sp.n.**

   Scutellum without markings ; hemelytron almost without markings, rarely with slight markings as in fig. 72..5

5. Second antennal segment black at distal extremity............*compositus*, **sp.n.**

   Second antennal segment not darkened at extremity...........*indecorus*, **sp.n.**

6. First antennal segment dark or black except at base, second nearly half dark or black distally (often appearing almost entirely dark unless cleared) ; hemelytron almost wholly dark (fig. 76).....................*ghesquierei*, Schout.

   First antennal segment not darkened ; second, if darkened at all, much less than half dark or black distally (occasionally appearing more than half dark unless cleared) ; hemelytron never so extensively darkened as in fig. 76.............7

7. Second antennal segment not or only very slightly darkened distally (*cf.* fig. 96)..8

   Second antennal segment dark or black distally......................................10

8. Hemelytron with slight markings as in figs. 53 and 54 ; length of insect more than 4·5 mm..*apicalis*, Fieb.

   Hemelytron without markings on corium or with merely a vague infuscation at distal end only of corium ; length of insect less than 4 mm......................9

9. Hemelytron without markings except a small but conspicuous dark spot at extreme distal end of corium and near inner corner of it, merging with infuscation on membrane (fig. 52) ; second antennal segment slightly darkened at distal end ; ♂ with laminate process on left paramere...............*virens*, **sp.n.**

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*All references to markings on the hemelytron are to those on that part of it which comprises the corium, clavus and embolium, not to those on the membrane.

Hemelytron usually entirely without markings (fig. 62) and if marked then marking consists merely of a vague infuscation at distal end of corium (fig. 63); second antennal segment not dusky distally; laminate process wanting..*palus*, **sp.n.**

10. Hemelytron with small hyaline spots in the dark markings (fig. 61) .... *notatus*, **sp.n.**

Hemelytron without such spots..11

11. Pronotum with a U-shaped black mark (rarely absent); corium entirely without markings..*nairobiensis*, Popp.

Pronotum without such mark, though sometimes dark throughout or otherwise marked; corium not entirely without markings.......................................12

12. Pronotum wholly or partly black..13

Pronotum not darkened..16

13. Hemelytron pale, nearly hyaline, with slight, inconspicuous markings only; a small species, length 3·5 mm. or less............................. *entadae*, **sp.n.**

Hemelytron with strong markings; relatively large species, length 4·3–4·9 mm... 14

14. Hemelytron with strong proximal and distal markings, but otherwise pale and nearly hyaline (fig. 82); scutellum conspicuously pale......... *thomasi*, Reut.

Hemelytron mainly dark; scutellum not conspicuously pale.....................15

15. Hemelytron with distal dark marking extending conspicuously to costal margin (fig. 73)..*tenebrosus*, **sp.n.**

Hemelytron rarely so, and then only vaguely (figs. 78, 79)...*chagweensis*, Popp.

16. Hemelytron markings as in figs. 67 and 68, not extending towards costal margin beyond an approximately median longitudinal line; second antennal segment much less than a quarter dark apically.................. *infirmus*, **sp.n.**

Hemelytron markings of different form and rarely thus confined; second antennal segment usually about a quarter dark apically........................17

17. Hemelytron markings as in fig. 75, slight and vague, the distal infuscation reaching costa and largely confined to costal half of hemelytron; a small species, length 3·5 mm. or less..*definitus*, **sp.n.**

Hemelytron markings different, usually more apparent and extensive, and usually extending from costa to inner margin, or if not, then absent in costal half of hemelytron; medium-sized species, length considerably more than 3·5 mm., usually about 4·0 mm...18

18. Hemelytron markings consisting principally of a single wedge-shaped streak and confined to inner half of hemelytron (fig. 74)..................*arboreus*, **sp.n.**

Hemelytron markings different, divided (though often incompletely) into proximal and distal portions, and extending into costal half of hemelytron, or if not divided, then consisting merely of a vague distal blotch (*cf.* figs. 55 and 48)..19

19. Thorax black or dusky ventrally..21

Thorax not so 20

20. Hemelytron markings consisting only of a dark blotch at distal end of corium, or, in rare cases, of a general darkening of inner half of hemelytron ; markings not extending towards costa (figs. 48, 49) .... *ricini*, **sp.n.**

Hemelytron markings consisting of proximal and distal blotches and extending to costa, but usually faint (figs. 64, 65) .... *simonyi*, Reut. (light forms)

21. Second antennal segment less than a quarter black distally 22

Second antennal segment black distally for a quarter of its length or slightly more 23

22. Cuneus, pronotum and scutellum light and bright in colour, contrasting conspicuously with darkly marked areas of hemelytron .... *cupressus*, **sp. n.**

Colour throughout much darker and more uniform, so that cuneus, in particular, does not contrast with darkly marked areas .... *complexus*, **sp.n.**

23. Second antennal segment with black portion terminating comparatively abruptly proximally (fig. 96)* .... *vosseleri*, Popp.

Second antennal segment with black portion shading off gradually proximally (fig. 96) .... *simonyi*, Reut. (dark forms)

Lygus vosseleri, Popp.

Sexes similar. Dorsally brownish, often with greenish tint, sometimes ochraceous ; ventrally similar but with an extensive dark brown or blackish area (fig. 83) on thorax extending on to propleura and abdomen, rarely extending back to form a lateral line on abdomen. Antennae with distal quarter of second segment black (fig. 96), the black portion terminating relatively abruptly proximally (not shading off gradually as in most others) ; third segment pale, dark at apex ; fourth dark, pale at base. Rostrum to apices of hind coxae. Hemelytron markings (fig. 50) usually slight and vague, consisting of a dark blotch at distal end of corium (usually more extensive than in *L. ricini*), tending to extend towards costal margin, and usually a faint oblique streak across proximal half of corium ; usually no spot in inner (obtuse) angle of clavus. Apex of cuneus black ; membrane infuscate with paler areas. Occasionally the markings are much more extensive, and the extreme form is shown in fig. 51. Distal half of hind femur slightly darker, sometimes slightly mottled.

Genitalia, figs. 3 and 29 ; laminate process large and very readily visible *in situ*.

Dimensions in mm. :—Length : ♂, 4·1 ; ♀, 4·2. Width of head : ♂ and ♀, 1·0. Width of pronotum : ♂, 1·5 ; ♀, 1·6.

Uganda, Kawanda, May 1944, 4 ♂♂, 11 ♀♀.

Very widely distributed in Africa.

This is the cotton *Lygus* of Uganda and other African countries. I have already listed and discussed its host plants in an earlier paper (1945). They include representatives of no less than 15 plant families, and the range of food plants of adults is very much wider. The host plants which, on account of their abundance, are the most important in the maintenance of a large population of this species in the countryside generally in Uganda are cotton, sorghum and several wild legumes.

This species, which has been called Brown Lygus and Sp. B in Uganda, was known as *L. simonyi*, Reut., from 1935 until the publication of my recent note (1947) showing that it is actually *L. vosseleri*, Popp., as it was believed to be prior to 1935.

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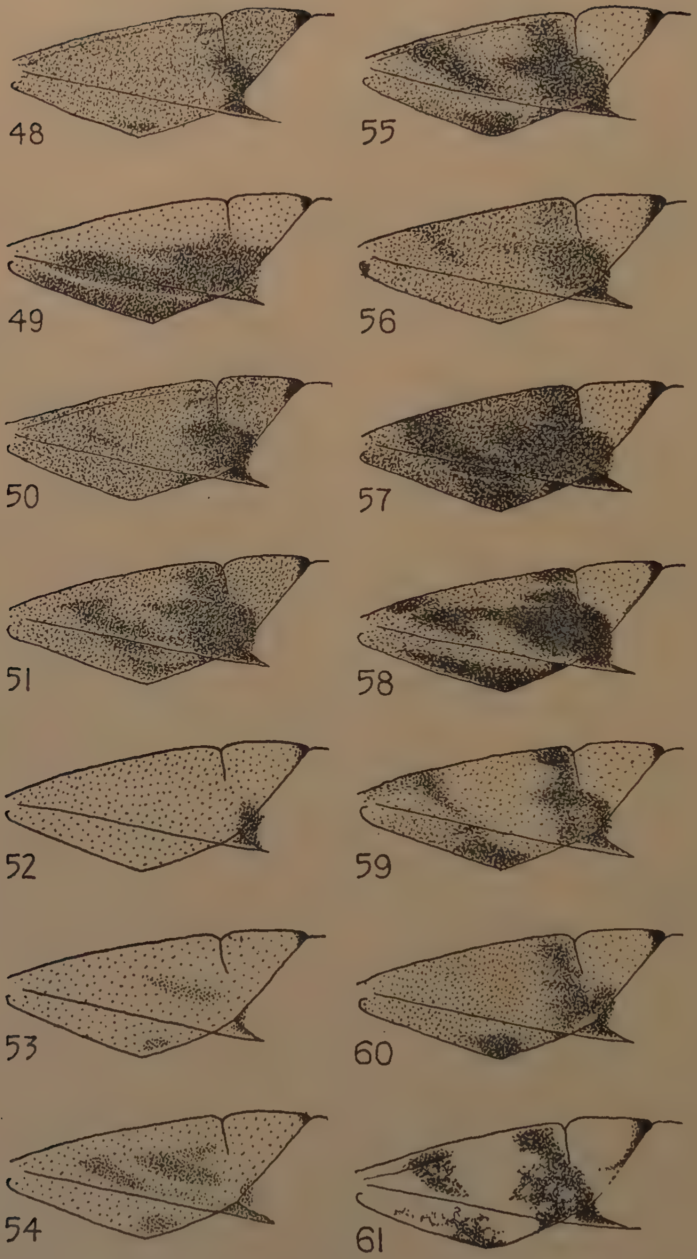
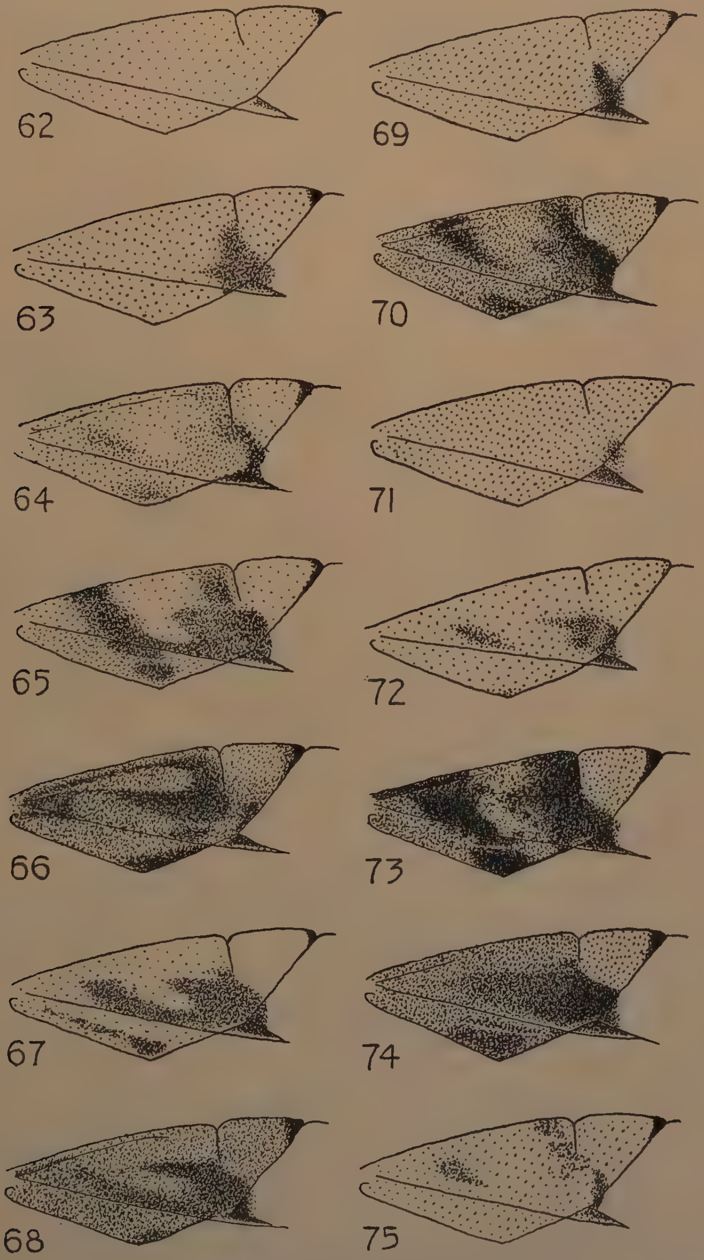
*This character is best judged with a hand lens, not a microscope.



Figs. 48–61.—Hemelytron of *Lygus* spp., drawn from cleared specimens to show markings, all somewhat diagrammatic and magnified to same size, not to same scale, to facilitate comparison ; markings on membrane omitted.—(48 and 49) *L. ricini*, sp.n. ; (50 and 51) *L. vosseleri*, Popp. ; (52) *L. virens*, sp. n. ; (53 and 54) *L. apicalis*, Fieb. ; (55–58) *L. cupressus*, sp.n. ; (59 and 60) *L. entadae*, sp.n. ; (61) *L. notatus*, sp.n.

Lygus ricini, sp.n.

Sexes similar. Dorsally green, sometimes yellowish-green or ochraceous; ventrally yellowish, often greenish-yellow and sometimes with vague pink markings; never black or dark beneath in either sex. Antennae with second segment black distally



Figs. 62–75.—See note for figs. 48–61.—(62 and 63) *L. palus*, sp.n.; (64 and 65) *L. simonyi*, Reut.; (66) *L. ugandanus*, sp.n.; (67 and 68) *L. infirmus*, sp.n.; (69) *L. nairobiensis*, Popp.; (70) *L. complexus*, sp.n.; (71 and 72) *L. compositus*, sp.n.; (73) *L. tenebrosus*, sp.n.; (74) *L. arboreus*, sp.n.; (75) *L. definitus*, sp.n.

for slightly less than a quarter of its length (fig. 96), third and fourth dark, the third slightly paler at base. Rostrum to apices of hind coxae. Hemelytron markings (fig. 48), usually slight and ill-defined (so that hemelytron appears nearly uniform in colour), consisting of a dark blotch at distal end of corium, not extending towards costal margin (*cf. L. vosseleri*, etc.), and a vague dark spot in inner (obtuse) angle of clavus; usually no oblique streak across proximal half of hemelytron. Apex of cuneus black; membrane infuscate with paler areas. Occasionally very dark specimens occur (fig. 49) but even in these the dark markings do not extend in front of an approximately median longitudinal line on that part which comprises the corium and embolium. Hind femur with dark mottling distally.

Genitalia, figs. 1, 2, and 28; figs. 1 and 2 show the laminate process in different degrees of compression and also illustrate variation in the teeth.

Dimensions in mm. :—Length: ♂, 3·9; ♀, 4·2. Width of head: ♂, 0·9; ♀, 1·0. Width of pronotum: ♂, 1·5; ♀, 1·6.

The type ♂ is of the green form with hemelytron markings as in fig. 48.

Uganda, Kawanda, May 1944, 4 ♂♂ (including type) and 6 ♀♀.

L. ricini (hitherto known in Uganda as Sp. A) breeds abundantly on *Ricinus communis* only, of the plants examined. It is certainly unusually restricted in its range of host plants, and the only other plant on which nymphs were found is sorghum. Adults are occasionally present on cotton, where they are liable to be confused with *L. vosseleri*, and on other plants of various families, but only as casual visitors.

It is highly probable that a specimen in the Berlin Museum recorded by Poppius under *L. vosseleri* as belonging to a species injurious to *Ricinus*, is actually *L. ricini*; I have not seen this specimen, which was taken at Amani, Tanganyika.

Lygus simonyi, Reut.

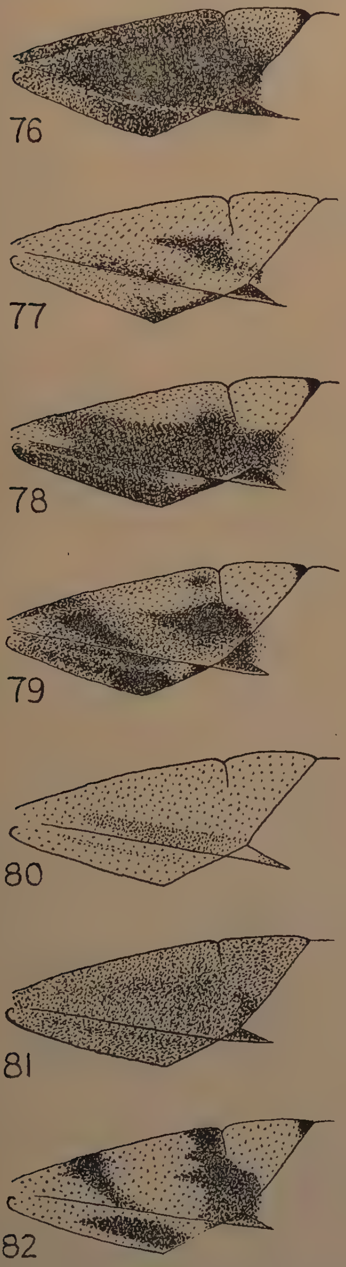
A rather small species. Green or yellowish-green, occasionally yellowish-brown. Both sexes usually pale ventrally, but ♂ often and ♀ rarely with a dusky or black area on thorax as in fig. 86; occasionally in both sexes this area extends as a vague lateral line on abdomen. Scutellum usually without markings, but occasionally with two longitudinal dark bands separated by a thin pale line and diverging posteriorly so as to leave apex conspicuously pale. Second antennal segment black at apex for rather more than a quarter of its length (fig. 96). Rostrum reaching very slightly beyond apices of hind coxae. Hemelytron usually nearly colourless and hyaline between markings, which are usually very faint (fig. 64) and consist of a distal blotch on corium extending almost to costa and connected near claval suture with a proximal oblique streak, and a vague spot on inner corner of clavus; in some specimens the markings are dark and conspicuous (fig. 65), but their form is nearly constant and the non-marked areas are nearly hyaline. Apex of cuneus black; membrane infuscate with pale areas. Hind femur darkly mottled distally, sometimes its distal two-thirds entirely dark brown.

Genitalia, figs. 14 and 15 (illustrating extent of variation in laminate process), 33, 94 and 95.

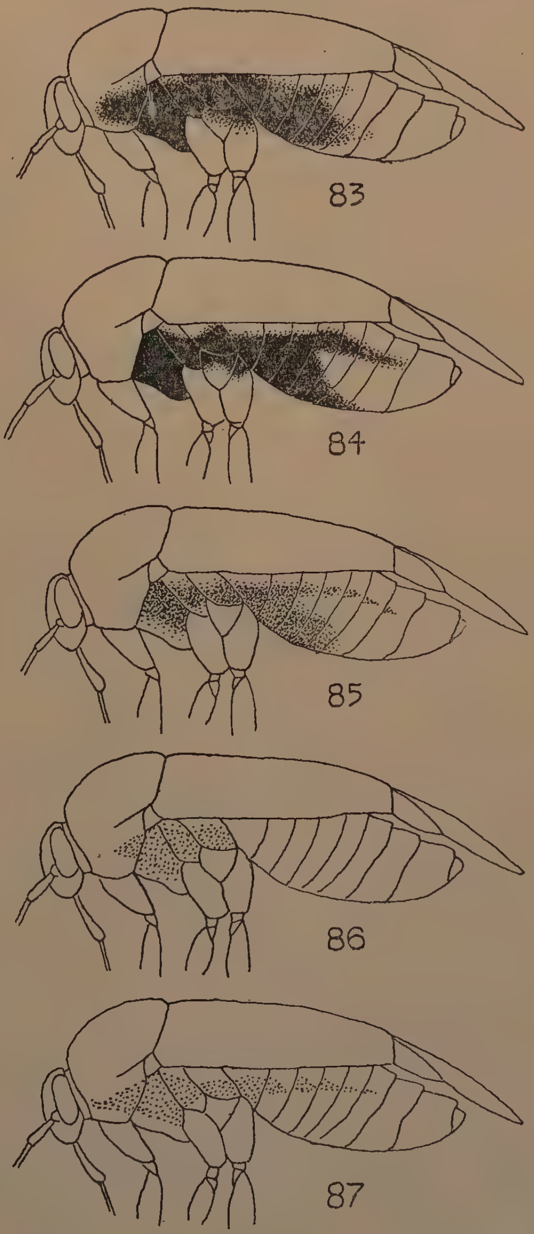
Dimensions in mm. :—Length: ♂ and ♀, 3·8. Width of head: ♂ and ♀, 1·0. Width of pronotum: ♂ and ♀, 1·5.

Uganda, Kawanda, May 1944, 14 ♂♂ and 7 ♀♀. Very widely distributed in Africa, from Aden to S. Africa.

L. simonyi, Reut., includes *L. schonlandi*, Dist. (Taylor, 1947). I have examined the three specimens which previously represented this species in the British Museum under the name *schonlandi*, Dist. Two (including the type) came from Grahamstown, S. Africa, and the third from Durban. They differ from almost all Uganda specimens and from the type of *simonyi*, Reut., from Aden in having the markings on the hemelytra, scutellum and ventral surface dark and conspicuous, but they agree well in other respects, notably in the details of the male genitalia. Distant's description



Figs. 76–82.—See note for figs. 48–61—(76) *L. ghesquierei*, Schout. ; (77) *L. vernoniae*, sp.n. ; (78 and 79) *L. chagweensis*, Popp. ; (80) *L. indistinctus*, sp.n. ; (81) *L. indecorus*, sp.n. ; (82) *L. thomasi*, Reut.



Figs. 83–87.—Lateral view of females of *Lygus* spp., diagrammatic, illustrating dark markings on ventral surface.—(83) *L. vosseleri*, Popp. ; (84) *L. arboreus*, sp.n. ; (85) *L. cupressus*, sp.n. ; (86) *L. simonyi*, Reut. ; (87) *L. infirmus*, sp.n.

is erroneous in two important respects. He stated that the base of the pronotum is piceous, implying that it has dark markings as in certain other species, but I have examined more than 100 specimens, including his type, and have never seen markings on the pronotum ; he obviously misinterpreted the translucency of the pronotum which gives certain parts of it a dark appearance. He also stated that the rostrum reaches " the intermediate coxae " ; actually it reaches slightly beyond the hind coxae and his error is due to the fact that in his specimens, which were gummed on cards, the heads were pressed forward in abnormal positions.

The host plants of *L. simonyi*, although covering a wide range, are very different from those of *L. vosseleri*, with which it has been confused, and do not include cotton or sorghum. The plants on which breeding is known to occur are *Amaranthus* sp., *Celosia* sp., *Rhus glaucescens*, *Cannabis sativa*, *Gynandropsis pentaphylla*, *Chenopodium* sp., *Combretum binderianum*, *Brassica schimperi*, *Erythrococca bongensis*, *Phaseolus* sp., *Cedrela* sp., *Melia azedarach* and *Theobroma* sp. This species is particularly abundant, as both nymphs and adults, on plants of the families Amarantaceae aud Chenopodiaceae and on *Gynandropsis pentaphylla*.

L. simonyi has previously been known in Uganda as Sp. H. The cotton Capsid was called *simonyi* until my recent paper (1947) pointed out that it is *vosseleri*.

Lygus cupressus, sp.n.

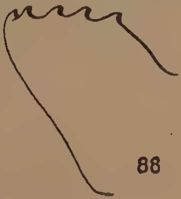
A very variable species. Dorsally, olive brown to ochraceous, occasionally almost entirely dark brown. Ventrally similarly coloured ; ♂ mainly dark brown, the dark area covering sternum, side of thorax and anterior part of abdomen, sometimes extending on to the propleura, peritreme and hind coxae, and extending backwards on abdomen as a lateral line ; ♀ ventrally dusky (fig. 85), almost invariably much less dark than ♂, the dusky area corresponding in extent to the dusky area of ♂, but in abnormally dark ♀♀ the underside resembles that of the normal ♂. Second antennal segment black distally for a little less than one-fifth of its length (fig. 96), third segment rather light in colour except distally, fourth dark. Rostrum long, reaching slightly beyond apices of hind coxae. Hemelytron markings very variable in depth of colour and extent, but general pattern constant (figs. 55–58, inclusive) and similar to that of extreme form of *L. vosseleri* (fig. 51) ; proximal oblique streak always present and distal blotch usually extending to costa and also extending medially towards base of hemelytron ; fig. 55 represents the normal form and figs. 56, 57 and 58 represent unusually pale, dark and darkly marked forms, respectively. Cuneus very conspicuous by reason of its paleness as compared with other parts of hemelytron ; apex of cuneus black. Membrane infuscate with pale areas. Hind femur dark brown and mottled except on basal third, fore and middle tibiae distally slightly darker and mottled.

Genitalia, figs. 11 and 32 ; laminate process large, very conspicuous *in situ*.

Dimensions in mm. :—Length : ♂, 4·1 ; ♀, 4·3. Width of head : ♂ and ♀, 1·0. Width of pronotum : ♂, 1·6 ; ♀, 1·7.

The type ♂ is olive brown and has the hemelytron markings of the normal form (fig. 55).

The great variation in colour and markings makes some specimens of this species difficult to determine on general appearance, but males are at once recognisable by the highly characteristic form of the laminate process on the left paramere, and both sexes can be distinguished from all other species known to me and with which they might be confused, except *L. complexus*, by the shortness of the dark portion of the second antennal segment. *L. cupressus* and *L. complexus* may be distinguished by their dimensions and by the colour of the third antennal segment which is mainly light in *cupressus* and mainly dark in *complexus*. This latter difference is constant in long series of both species, but is not easily observed unless the antennae are cleared and mounted on microscope slides. An additional means of distinguishing these two species is afforded by the length of the antennae, which are shorter, relative to head and body dimensions, in *cupressus* than *complexus*. This is best demonstrated by comparing the length of the second antennal segment with the width of the head or pronotum. Measurement of six specimens of each sex of each species gave the following ratios :—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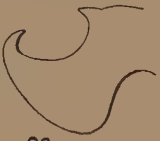
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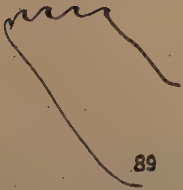
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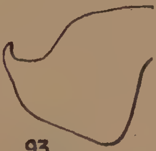
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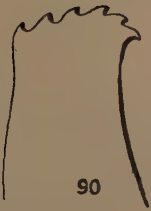
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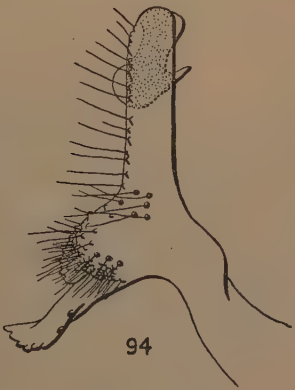
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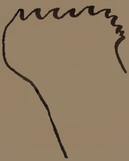
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Figs. 88–91.—Laminate process of left paramere of (88 and 89) *L. indecorus*, sp.n., × 150 ; (90) *L. thomasi*, Reut., × 235 ; (91) *L. indistinctus*, sp.n., × 150.

Figs. 92 and 93.—Hooked process of left paramere of (92) *L. indistinctus*, sp.n., and (93) *L. indecorus*, sp.n.

Fig. 94.—Left paramere of *L. simonyi*, Reut. (*cf.* fig. 95).

	♂		♀	
	cupressus	*complexus*	*cupressus*	*complexus*
2nd antennal segment : width of head ...	1 : 0·65	1 : 0·57	1 : 0·67	1 : 0·59
2nd antennal segment : width of pronotum	1 : 1·00	1 : 0·86	1 : 1·07	1 : 0·95

L. cupressus has a wide range of host plants, including trees and herbaceous plants. It is known to breed on *Vernonia* spp., *Cupressus* sp., *Aberia gardneri* and *Buddleia polystachya*. Hitherto it has been referred to in Uganda as Sp. E.

Lygus complexus, sp.n.

Brownish-yellow or brown. Dark markings ventrally as described and figured for *L. cupressus* (both sexes), and similarly variable, but less conspicuous because ground colour darker. Antennae with second segment black distally for one-fifth of its length (fig. 96), third mainly dark, fourth dark. Rostrum long, reaching slightly beyond apices of hind coxae. Hemelytron markings (fig. 70) as in *L. cupressus*. Cuneus not conspicuously pale (*cf. L. cupressus*), its apex black; membrane infuscate with pale areas. Distal two-thirds of hind tibia dark brown, sometimes mottled.

Genitalia, figs. 17 and 38; laminate process small.

Dimensions in mm.:—Length: ♂, 4·0; ♀, 3·8. Width of head: ♂ and ♀, 0·9. Width of pronotum: ♂, 1·4; ♀, 1·5.

UGANDA, Kawanda, May, 1944, 10 ♂♂ (including type) and 10 ♀♀.

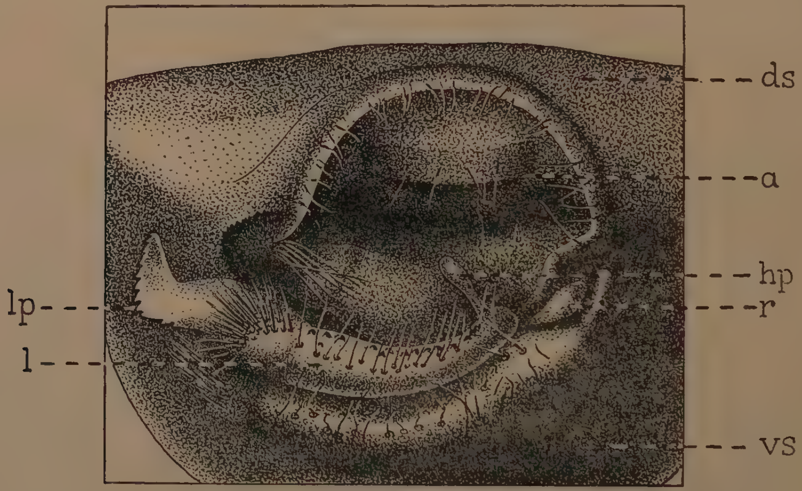


Fig. 95.—Posterior view of abdomen of male of *L. simonyi*, Reut., to show, in particular, the normal position and appearance of the left paramere and its appendages in the *simonyi* group of species.—a, anal aperture; ds, dorsal surface of abdomen; hp, hooked process (edge view) of left paramere; l, left paramere; lp, laminate process of left paramere; r, right paramere; vs. ventral surface of abdomen.

Males of this species are at once distinguishable from those of *L. cupressus*, with which they might otherwise be confused, by the laminate process on the left paramere, which is readily visible *in situ*. Other distinguishing features, which apply to both sexes, are described under *L. cupressus*. It should be noted also that *L. complexus* is much duller in general colour and that its cuneus, though coloured similarly to that of *L. cupressus*, is not conspicuously pale by contrast with other parts of the hemelytron.

The range of host plants of *L. complexus* is extremely limited, the only one known being *Cupressus* sp. Adults have been taken on *Cinnamomum* sp. in large numbers, and on *Aberia gardneri* and *Persea gratissima* in small numbers, but there is no evidence of breeding on these trees.

L. complexus has hitherto been called Sp. N in Uganda.

Lygus entadae, sp.n.

A small species, readily recognised by its contrasting markings. Greenish-yellow, sometimes brownish-yellow. Pronotum in ♂ black; in ♀ greenish-yellow anteriorly and dark brown posteriorly, the dark brown portion being in the form of a broad transverse band. Scutellum greenish-yellow or pale green, conspicuous. Ventral surface pale greenish-yellow, but thorax black laterally in ♂, the black area including propleura but not extending to coxae, peritreme or abdomen; female wholly pale ventrally. Second antennal segment black distally for a little less than a quarter of its length (fig. 96). Rostrum to apices of hind coxae. Hemelytra almost colourless and semi-transparent so that its markings are usually conspicuous by contrast; markings (figs. 59 and 60) consisting of a dark blotch at distal end of corium, extending to costa, a dark, ill-defined spot in inner (obtuse) angle of clavus, and a proximal oblique streak on corium which is present in ♂ only. Apex of cuneus black; membrane infuscate with pale areas. Distal half of hind femur darker and mottled.

Genitalia, figs. 12 and 43.

Dimensions in mm. :—Length: ♂ 3·3; ♀ 3·5. Width of head: ♂ and ♀, 0·9. Width of pronotum: ♂, 1·2; ♀, 1·4.

UGANDA, Kawanda, May 1944, 6 ♂♂ (including type) and 9 ♀♀.

The only known host plant is *Entada abyssinica*, and since nymphs were never obtained from any herbaceous plant it is probable that the range of host plants is limited to certain flowering trees.

This species has been called Sp. F in Uganda.

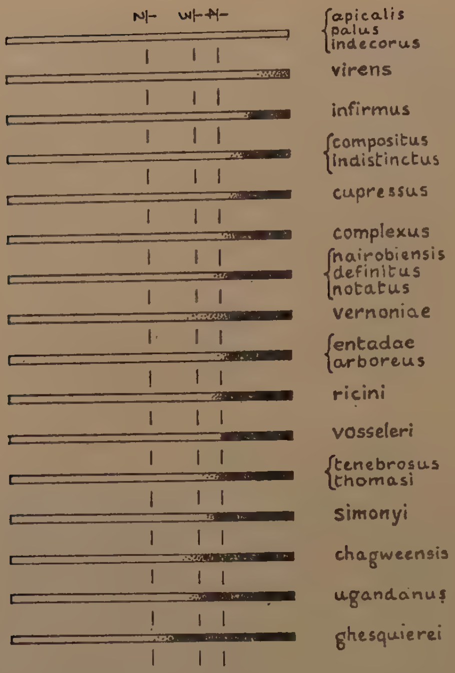
Lygus thomasi, Reut.

Sexes similar. Pale green or yellowish green, with exceptionally conspicuous dark markings. Antennae with distal quarter of second segment black (fig. 96). Rostrum reaching apices of hind coxae. Pronotum with a dark brown or black transverse band posteriorly but extreme posterior margin pale. Hemelytron very pale and nearly transparent, with strong, dark brown markings (fig. 82) consisting of a blotch at distal end of corium extending from costa to inner margin but darkest near costa, a proximal oblique streak extending from costa to claval suture and a longitudinal blotch on clavus; apex of clavus and of cuneus black; membrane darkly infuscate with pale areas. Thorax dark or black ventrally and laterally, except coxae, peritreme and propleura, the dark area usually extending posteriorly as a vague lateral line on abdomen. Scutellum conspicuously pale by contrast with surrounding markings. Distal half of hind femur darkly mottled.

Genitalia, fig. 90.

Dimensions in mm. :—Length: ♂, 4·8; ♀, 4·6. Width of head: ♂ and ♀, 1·0. Width of pronotum: ♂, 1·65; ♀, 1·75.

I have redescribed this species, which I have never obtained in the field, from seven specimens in the Paris and Helsinki Musems. It is not represented in the British Museum and I have not been able to see the type, but since the Helsinki specimens were determined by Poppius and the Paris ones by Sjöstedt I accept the determinations. There has been a tendency, due to the inadequacy of the original description, to identify other species in collections as *thomasi*; actually, it is a very distinctive species. The specimens which I have examined were all collected by Sjöstedt at Kibonoto on Mt. Kilimandjaro in 1905–6. They comprise 3 ♂♂ and 4 ♀♀. My drawings of the toothed process of the left paramere, the hemelytron and the second antennal segment were prepared without removing these parts from the specimens as I was anxious to avoid mutilation; for this reason I have been unable to examine and illustrate the hook of the paramere.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notatus
vernoniae
entadae
arboreus
ricini
vosseleri
tenebrosus
thomasi
simonyi
chagweensis
ugandanus
ghesquierei

Fig. 96. Second antennal segment of *Lygus* spp., diagrammatic, to show length of dark portion relative to length of segment.

Lygus tenebrosus, sp.n.

A rather large, dark species, easily recognised. Sexes similar. Dorsally brown, ventrally brownish-black in both sexes (except peritreme which is usually pale), but in the female the abdomen is occasionally brown. Face variable, mainly dark brown or black, less dark along margins of eyes and on vertex. Second antennal segment with distal quarter black (fig. 96). Rostrum to apices of hind coxae. Pronotum usually with posterior half black, sometimes less than half black, often wholly black; extreme posterior margin always pale; calli more or less black, often forming a pair of conspicuous black spots. Scutellum usually with two broad, longitudinal, black bands, widening posteriorly; apex and anterior angles brown. Hemelytron markings (fig. 73) very dark, nearly black, sometimes more extensive than in figure so that hemelytron almost wholly dark except cuneus and membrane; markings similar to those of *vosseleri*, *cupressus* and *complexus*, but much more pronounced. Apex of cuneus black; membrane darkly infuscate with pale areas. Hind tibia dark brown or black except on proximal third, sometimes somewhat mottled distally; other tibiae usually darkly mottled on distal two-thirds.

Genitalia, figs. 18 and 40.

Dimensions in mm. :—Length : ♂, 4·3 ; ♀, 4·5. Width of head : ♂, 1·0 ; ♀, 1·1. Width of pronotum : ♂, 1·6 ; ♀, 1·7.

In the type ♂ the face is dark brown except laterally and above, the pronotum has the calli conspicuously black and a broad transverse black band posteriorly, the scutellum has the dark bands so widened except anteriorly that they meet medially and are almost triangular, the hemelytron markings are similar to fig. 73 but rather more extensive, and the hind tibiae are mainly black.

UGANDA, Kawanda, May 1944, 8 ♂♂ (including type) and 7 ♀♀.

The only known host plants of this species, which has previously been called Sp. P in Uganda, are *Phytolacca dodecandra* and *Capsicum* sp.; it is a relatively rare and local insect in Uganda.

Lygus chagweensis, Popp.

A rather large, dark brown species. Head brown, tylus and sometimes frons black. Second antennal segment (fig. 96) nearly one-third black distally (but length of black portion difficult to judge in dark specimens unless cleared). Rostrum to apices of hind coxae. Pronotum in ♂ usually wholly black, in ♀ usually brown with a pair of black spots anteriorly (on calli) and a broad, black, transverse band posteriorly; occasionally wholly black in ♀ also; extreme posterior margin always pale. Scutellum usually nearly black in ♂ except for a diamond-shaped area at apex; brown without markings in ♀. Ventrally, ♂ very dark brown or black, including propleura but excepting coxae and peritreme which are pale and central area of abdomen which is usually less dark than sides, though abdomen is sometimes wholly black; ♀ ventrally mainly brown, sides of thorax dark brown or blackish; in both sexes pale forms occur and in these there is a conspicuous dark lateral line on abdomen, which is otherwise pale, but sides of thorax always dark. Hemelytron markings very dark and extensive, usually as in fig. 78, corium being wholly dark brown, except at extreme base and distally near embolium, and clavus almost wholly dark; but frequently markings paler in centre of corium (fig. 79) so as to be divided into a distal blotch and a proximal oblique streak similar to but darker than the normal markings of certain other species (*e.g.*, *cupressus*, fig. 55), and occasionally whole hemelytron to cuneal fracture nearly black except for a short length of distal half of embolium; forms intermediate between those figured are common. Cuneus with black spot at apex; membrane darkly infuscate with pale areas. Hind tibia with distal half or two-thirds dark brown, often somewhat mottled, particularly in ♀.

Genitalia, figs. 27 and 46; laminate process very small.

Dimensions in mm.:—Length: ♂, 4·7; ♀, 4·9. Width of head: ♂, 1·0; ♀, 1·1. Width of pronotum: ♂, 1·6; ♀, 1·8.

This is a forest species which breeds on *Basella alba*. It has been referred to previously as Sp. T in Uganda.

Lygus infirmus, sp.n.

Dorsally yellowish-green, occasionally olive-brown; ventrally greenish-yellow to greenish-grey. ♀ usually without appreciable darkening ventrally, but sometimes with slight darkening to the extent shown in fig. 87; ♂ dusky or dark brown in areas shown for ♀ in fig. 87, always darker than ♀, the dark area extending on to abdomen as a distinct lateral line. Second antennal segment (fig. 96) black distally, the black portion being much less than a fifth of length of segment (and less, relative to length of segment, than in any other species here described). Rostrum long, reaching slightly but noticeably beyond apices of hind coxae. Scutellum with two dark, longitudinal bands, but these sometimes very faint. Hemelytron markings (figs. 67 and 68) characteristic, consisting of a dark blotch distally on corium connected along posterior border of corium with a proximal oblique streak, and a vague dark spot in inner angle of clavus; markings not extending in front of an approximately median longitudinal line on corium (except very rarely in specimens in which the corium is almost wholly dark). Apex of cuneus black; membrane infuscate with pale areas. Hind tibia dark distally for about two-thirds length and somewhat mottled.

Genitalia, figs. 22 and 23, which illustrate variation in laminate process, and 36.

Dimensions in mm.:—Length: ♂, 3·9; ♀, 4·0. Width of head: ♂ and ♀, 1·0. Width of pronotum: ♂, 1·4; ♀, 1·5.

The type ♂ has hemelytron markings as in fig. 67 and the two bands on the scutellum distinct.

UGANDA, Kawanda, May 1944, 11 ♂♂ (including type) and 4 ♀♀.

The host plants appear to be restricted to certain legumes ; of the great variety of plants examined, only *Caesalpinia sepiaria* and *Cajanus indicus* are host plants, but on both of these breeding occurs and nymphs and adults are plentiful. Adults have been taken in small numbers on *Hoslundia opposita* and *Pseudarthria* sp.

This species has hitherto been known as Sp. L in Uganda.

Lygus definitus, sp.n.

A small species. Green to pale yellowish-brown, ♂ darker than ♀ ; ♀ pale throughout, ♂ slightly dusky ventrally on sternum, sides of thorax except peritreme, and usually on first visible abdominal segment. Second antennal segment (fig. 96) black distally for slightly less than a quarter of its length. Rostrum reaching very slightly beyond apices of hind coxae. Hemelytron markings (fig. 75) very slight, consisting of a vague dark blotch at distal end of corium and embolium, extending to costal (embolar) margin but not to claval suture, and a more or less wedge-shaped streak in proximal half of corium rarely reaching costal margin or claval suture. Cuneus with black spot at apex ; membrane infuscate with pale areas. Distal two-thirds of hind tibia, and usually distal third of other tibiae, darkly mottled, but never continuously dark.

Genitalia, figs. 24 and 25, illustrating variation in laminate process, and 42 ; the form of the process is highly characteristic.

Dimensions in mm. :—Length : ♂, 3·4 ; ♀, 3·5. Width of head : ♂ and ♀, 0·9. Width of pronotum : ♂ and ♀, 1·3.

UGANDA, Kawanda, May 1944, 10 ♂♂ (including type) and 5 ♀♀.

L. definitus (called Sp. R in Uganda hitherto) breeds abundantly on *Sida schimperiana* and *Celosia* sp., but was not obtained from any other plant.

Lygus arboreus, sp.n.

Dorsally olive-brown, ventrally similar but with extensive dark brown or blackish areas in both sexes, darker and more extensive in ♂ than in ♀ (fig. 84, extreme form of ♀) ; dark area not extending to propleura but extending on to abdomen and backwards on abdomen as a lateral line ; in ♂, abdomen usually mainly dark brown, except posterior third (the large genital segment) and above lateral dark line. Second antennal segment black distally for slightly less than a quarter of its length (fig. 96). Rostrum reaching very slightly beyond apices of hind coxae. Hemelytron markings as in fig. 74 ; dark area on corium not divided and not extending towards costal margin beyond a nearly median, longitudinal line ; markings often much fainter than in figure. Apex of cuneus black ; membrane infuscate with pale area. Hind tibia usually with distal two-thirds slightly darkly mottled, occasionally wholly dark except proximal third.

Genitalia, figs. 20 and 21 (showing laminate process of two specimens in different degrees of compression) and 41.

Dimensions in mm. :—Length : ♂, 4·1 : ♀, 4·4. Width of head : ♂ and ♀, 1·0. Width of pronotum : ♂, 1·6 ; ♀, 1·7.

The type ♂ has the hemelytron markings as in fig. 74 and the dark markings on the abdomen almost as shown in fig. 84 for the ♀.

UGANDA, Kawanda, May 1944, 11 ♂♂ (including type) and 6 ♀♀.

The adults of this species mainly frequent flowering trees and shrubs, including coffee. The known host plants are *Persea gratissima*, *Caesalpinia sepiaria* and *Cannabis sativa*.

L. arboreus has been called Sp. Q in Uganda.

Lygus notatus, sp.n.

A rather small species. Greenish-yellow. Ventrally, ♂ partly dark brown or dusky, dark area covering sides of thorax but not extending to propleura, and abdomen usually with a vague dark lateral line, but otherwise pale ; ♀ entirely pale beneath. Scutellum with a pair of dark triangular marks dividing it so that its apex and its two basal angles are pale. Second antennal segment black distally, the black portion being a little less than a quarter of the length of the segment (fig. 96). Rostrum to apices of hind coxae. Hemelytron markings (fig. 61) conspicuous because non-marked areas almost hyaline ; markings consist of a distal dark blotch on corium extending to costa, a proximal oblique streak, and an irregular darkening at inner angle of clavus ; in all the hemelytron markings there are small irregular hyaline patches which give them a mottled appearance and are highly characteristic. Apex of cuneus black ; membrane slightly infuscate with pale areas. Distal half or third of hind femur darkly mottled.

Genitalia, figs. 16 and 34.

Dimensions in mm. :—Length : ♂, 3·6 ; ♀, 3·8. Width of head : ♂ and ♀, 1·0. Width of pronotum : ♂, 1·5 ; ♀, 1·6.

UGANDA, Kawanda, May 1944, 4 ♂♂ (including type) and 8 ♀♀.

Adults of *L. notatus*, which is relatively uncommon, have been taken on several different plants, including cotton, but the only known host plant is *Combretum* sp. This insect has previously been called Sp. J in Uganda.

Lygus nairobiensis, Popp.

Sexes similar. Ochraceous to pale green ; ventrally pale throughout. Antennae with second segment black distally for nearly a quarter of its length (fig. 96). Rostrum long, reaching slightly beyond apices of hind coxae. Pronotum with two broad, longitudinal, brownish-black bands, very conspicuous, well separated anteriorly but curving inwards posteriorly and almost or quite meeting near posterior margin, which they do not quite reach ; black bands rarely absent, and occasionally developed to such a degree that pronotum is almost entirely black except along posterior margin. Scutellum always pale, without markings. Hemelytron without markings except for small dark blotch shown in fig. 69. Apex of cuneus black ; membrane infuscate with pale areas. Distal quarter of hind tibia usually darkly mottled.

Genitalia, figs. 13 and 37.

Dimensions in mm. :—Length : ♂, 4·2 ; ♀, 4·4. Width of head : ♂, 0·9 ; ♀, 1·0. Width of pronotum : ♂, 1·5 ; ♀, 1·6.

UGANDA, Kawanda, May 1944, 8 ♂♂ and 7 ♀♀. Widely distributed in E. Africa.

I have identified this species from the original description of Poppius ; it is a very distinctive species and I have no doubt as to its identity. It has been referred to as Sp. M in Uganda.

The host plants are herbaceous and include *Vernonia* spp., *Hoslundia opposita*, *Ocimum* sp., *Cajanus indicus*, *Cannabis sativa* and *Triumfetta* sp. Adults occasionally visit cotton, but no breeding occurs on it.

Lygus virens, sp.n.

A rather small, pale species. Sexes similar. Green throughout. Second antennal segment slightly dusky at extremity (fig. 96). Rostrum to apices of hind coxae. Hemelytron always without markings except for dark spot shown in fig. 52 and the usual infuscation, with pale areas, on the membrane ; apex of cuneus black. Hind femur usually somewhat darker distally, the darker portion slightly mottled.

Genitalia, figs. 5–9 (showing range of variation in laminate process, which is minute but of highly characteristic form) and 30.

Dimensions in mm. :—Length : ♂, 3·6 ; ♀, 3·8. Width of head : ♂ and ♀, 0·9. Width of pronotum : ♂ and ♀, 1·4.

Uganda, Kawanda, May 1944, 9 ♂♂ (including type) and 4 ♀♀.

L. virens (hitherto known as Green Lygus or Sp. C in Uganda) breeds abundantly on sorghum and on several trees having dense inflorescences of minute flowers, notably *Entada abyssinica*, *Cedrela* sp. and probably *Mangifera indica*, on the flowers of which adults are plentiful. Cotton is also a host plant, but rarely.

Lygus indistinctus, sp.n.

A green species, almost entirely devoid of markings both dorsally and ventrally. Sexes similar. Second antennal segment (fig. 96) dark (not black) at apex for about one-fifth (always less than a quarter) of its length. Rostrum short, reaching to apex of middle coxae only, differing in this respect from that of all other species dealt with here. Hemelytron with a faint darkening along claval margin of corium (sometimes difficult to detect) as in fig. 80 ; otherwise without markings. Cuneus not black at apex, thus resembling that of *compositus*, *indecorus* and *vernoniae* only ; membrane usually entirely hyaline, rarely infuscate with pale areas. Hind tibia pale throughout or only very slightly darkened distally.

Genitalia ; figs. 91 and 92.

Dimensions in mm. :—Length : ♂, 4·6 ; ♀, 4·5. Width of head : ♂ and ♀, 1·0. Width of pronotum : ♂ and ♀, 1·6.

The type ♂ has the hemelytron markings as figured and the membrane hyaline.

Uganda, Kawanda, May 1944, 6 ♂♂ (including type) and 9 ♀♀.

The only known host plant of this species, which has hitherto been known as Sp. V, is bracken.

Lygus vernoniae, sp.n.

A rather large, elongate species. Green to yellowish-green. Ventrally pale in both sexes, but some ♂♂ have sides of thorax dark and a dark lateral line on abdomen. Second antennal segment (fig. 96) with distal quarter, or slightly less, black, the black portion usually shading off gradually proximally so that its length is difficult to estimate except in cleared specimens. Rostrum to apices of hind coxae. Pronotum with a pair of dark brown spots anteriorly (on the calli), which are larger in most ♂♂ than in most ♀♀, and a dark brown transverse band posteriorly, wider and darker in most ♂♂ than in most ♀♀, occasionally absent in ♀. Scutellum with a pair of longitudinal dark lines, approximating anteriorly and diverging posteriorly, sometimes faint or absent except anteriorly and more distinct in most ♂♂ than in most ♀♀. Hemelytron markings (fig. 77) consisting of a wedge-shaped median streak distally on corium, a vague darkening along claval margin on corium (often almost imperceptible) and another vague darkening at inner angle of clavus ; no oblique streak across proximal end of corium. Cuneus not black at apex, differing in this respect from that of all other species dealt with here except *compositus*, *indistinctus* and *indecorus* ; membrane infuscate with pale areas. Hind tibia darkly mottled in distal half, particularly in ♂, mottling scarcely perceptible in ♀.

Genitalia, figs. 26 and 45.

Dimensions in mm. :—Length : ♂, 4·6 ; ♀, 5·2. Width of head : ♂ and ♀, 1·1. Width of pronotum : ♂, 1·7 ; ♀, 1·9.

The type ♂ has the scutellum without markings except anteriorly, hemelytron markings as in fig. 77, and hind tibia only slightly darkened distally.

UGANDA, Kawanda, May 1944, 10 ♂♂ (including type) and 5 ♀♀.

This species breeds abundantly on a species of *Vernonia*, but was not obtained from any other plant. It has been called Sp. S in Uganda hitherto.

Lygus compositus, sp.n.

Sexes similar. Yellowish-brown, yellowish-green or green, throughout. Second antennal segment black distally for $\frac{1}{6}$–$\frac{1}{5}$ of its length (fig. 96). Rostrum long, reaching slightly but obviously beyond apices of hind coxae. Hemelytron usually almost entirely without markings (fig. 71), but some males have a dark blotch at distal end of corium and an oblique streak proximally, both confined to that half of the corium which adjoins clavus (fig. 72). Cuneus without black spot at apex (though sometimes slightly dark apically), differing in this respect from all other species dealt with here except *vernoniae*, *indistinctus* and *indecorus* ; membrane infuscate with pale areas. Hind tibia usually with slight dark mottling distally.

Genitalia, figs. 4 and 44 ; laminate process small, not easily seen *in situ*.

Dimensions in mm. :—Length : ♂, 4·5 ; ♀, 4·6. Width of head : ♂ and ♀, 1·0. Width of pronotum : ♂, 1·6 ; ♀, 1·7.

The type ♂ has a trace of the markings shown in fig. 72.

UGANDA, Kawanda, May 1944, 8 ♂♂ (including type) and 12 ♀♀.

L. compositus (hitherto known as Sp. O in Uganda) breeds on *Vernonia* sp., *Hibiscus* sp. ? *macranthus* and *Triumfetta* sp.

Lygus indecorus, sp.n.

A green species, almost devoid of markings dorsally and ventrally. Sexes similar. Second antennal segment (fig. 96) not darkened at apex. Rostrum reaching to or very slightly beyond apices of hind coxae. Hemelytron (fig. 81) without markings, except a vague darkening at distal end of corium. Cuneus not black at apex, thus resembling that of *indistinctus*, *compositus* and *vernoniae* only ; membrane infuscate with pale areas. Hind tibia not or only very slightly darkened distally.

Genitalia, figs. 88, 89 and 93.

Dimensions in mm. :—Length : ♂, 5·0 ; ♀, 5·5. Width of head : ♂, 1·06 ; ♀ 1·1. Width of pronotum : ♂, 1·7 ; ♀, 1·8.

KENYA, Ruiru, Eldama and Mt. Elgon, 1932, 9 ♂♂ (including type) and 8 ♀♀.

These specimens were collected on coffee and *Lantana* by H. C. James, but it is not known whether these are true host plants. I have never collected this species.

Lygus ghesquierei, Schout.

A rather large, reddish-brown species. Ventrally, ♂ dusky or blackish except on propleuron, middle area of sternum, peritreme and middle segments of abdomen ; ♀ wholly pale ventrally. First antennal segment dark brown or black, differing in this respect from that of all other species dealt with here ; second segment (fig. 96) nearly half black or dark brown distally (appearing almost entirely dark in dark specimens until cleared), the dark portion shading very gradually into the paler proximal portion. Rostrum to apices of hind coxae. Hemelytron (fig. 76) almost wholly dark, usually slightly paler proximally; embolium and embolar region of corium often slightly paler than remainder of corium. Cuneus with black spot at apex; membrane darkly infuscate with paler areas. Hind tibia sometimes uniformly dark brown throughout except at base, sometimes merely darkly mottled except in basal third.

Genitalia, figs. 19 and 47.

Dimensions in mm. :—Length : ♂, 4·6 ; ♀ 4·7. Width of head : ♂ and ♀, 1·0. Width of pronotum : ♂, 1·7 ; ♀, 1·8.

UGANDA, various localities and dates, 6 ♂♂ and 12 ♀♀ (*H. Hargreaves*).

The only known host plant of this insect, which is probably a forest species and has hitherto been called Sp. U in Uganda, is coffee.

This species has often been confused in collections with *L. arboreus*, partly because they occur together on coffee trees (with *L. coffeae*, China), but the two species are readily distinguishable not only by the processes of the left paramere, which are very different, but also, and in both sexes, by the colour of the first antennal segment, the length of the dark portion of the second antennal segment, and the hemelytron markings (which, however, cannot be judged satisfactorily unless the hemelytra are either raised from the body or, preferably, cleared and mounted on a microscope slide).

Lygus apicalis, Fieb.

A rather large, elongate species. Green throughout, commonly tending to yellowish, usually pale; underside wholly pale. Antennae, including extremity of second segment (fig. 96), pale, fourth segment slightly darker. Rostrum long, reaching slightly beyond apices of hind coxae. Hemelytron usually with very slight markings as in fig. 53, occasionally more extensively marked on corium as in fig. 54, and often without markings. Extreme apex of cuneus black or dark; membrane infuscate with large pale areas. Hind femur distally somewhat darker and mottled.

Genitalia, figs. 10 and 31.

Dimensions in mm. :—Length : ♂, 4·6 ; ♀, 4·7. Width of head : ♂ and ♀, 1·0. Width of pronotum : ♂, 1·6 ; ♀, 1·7.

UGANDA, Kawanda, May 1944, 5 ♂♂ and 8 ♀♀.

The known host plants of the species, which are mainly composites and include several common weeds, are *Conyza* sp., *Erigeron linifolius*, *Microglossa* sp., *Senecio* spp., *Vernonia* spp., *Hoslundia opposita*, *Coriandrum sativum* and *Lantana* sp. It occurs not only in Africa but also in North and Central America and in Europe.

L. apicalis has previously been known in Uganda as Sp. D.

Lygus ugandanus, sp.n.

Sexes similar. Brown dorsally and ventrally, without black or dark markings ventrally, but sometimes abdomen slightly mottled. Head light brown, tylus dark brown or black near extremity. Second antennal segment with distal third black (fig. 96). Rostrum reaching beyond middle coxae but not to apex of hind coxae. Hemelytron markings (fig. 66) in the form of a loop, highly characteristic. Cuneus with black spot at apex; membrane infuscate with pale areas. Hind tibia usually dark brown on proximal half and with dark mottling near apex.

Genitalia, fig. 35; laminate process wanting.

Dimensions in mm. :—Length : ♂ and ♀, 4·0. Width of head : ♂, 1·1 ; ♀, 1·0. Width of pronotum : ♂, 1·5 ; ♀, 1·6.

Uganda, Serere, July 1940, and July and September 1942, 4 ♂♂ (including type) and 5 ♀♀.

This species (previously called Sp. K in Uganda) appears to be rare, but this may mean merely that its principal habitat and host plant are unknown. The nine specimens on which this description is based are the only ones known to me. Adults have been collected on *Combretum* sp., *Bridelia micrantha* and *Morus* sp., and the rather scanty evidence available suggests that this insect is restricted to the short-grass country and breeds on certain trees.

Lygus palus, sp.n.

A very small species. Pale brownish-yellow or pale green ; ♀ pale throughout, ♂ usually pale throughout but sometimes with pronotum darkly pigmented posteriorly. Second antennal segment not darker at apex (fig. 96). Rostrum to apices of hind coxae. Hemelytron often without markings (fig. 62), except a black spot at apex of cuneus and infuscation with hyaline areas on membrane, but some specimens (particularly ♂♂ having a dark pronotum, but occasionally ♀♀ also) have a vague infuscation at distal end of corium, not extending to costa (fig. 63, which represents the extreme form). Hind femur darkly mottled distally.

Genitalia : laminate process wanting ; terminal hook of left paramere with an erect process as in fig. 39.

Dimensions in mm. :—Length : ♂, 3·2 ; ♀, 3·5. Width of head : ♂ and ♀, 0·9. Width of pronotum : ♂, 1·2 ; ♀, 1·3.

Type ♂ has pronotum dark except antero-medially and slight infuscation at end of corium similar to fig. 63.

Uganda, various localities, 1942–1944, 5 ♂♂ (including type) and 6 ♀♀.

This species, regardless of the colour of the pronotum, is readily distinguished from all others dealt with here by its small size, the absence of black or dark pigments at the apex of the second antennal segment, the complete or almost complete absence of markings on the hemelytron, and by the male genitalia.

L. palus appears to be associated principally with flowering trees. It is known to breed on *Anacardium occidentale, Mangifera indica, Caesalpinia sepiaria, Entada abyssinica* and *Triumfetta* sp.

Hitherto this species has been referred to in Uganda as Sp. G.

References.

The principal works, other than original descriptions, which I have consulted or to which I have referred in the text are :—

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Taylor, T. H. C. (1945). *Lygus simonyi,* Reut., as a Cotton Pest in Uganda.—Bull. ent. Res., **36,** p. 121.

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THE IDENTITY OF *PHALAENA CHRYSORRHOEA*, LINNAEUS, 1758.

By C. L. Collenette, F.R.E.S.

E.M.N

Rothschild in 1917 (Novit. Zool., **24**, p. 355) stated that the above name, which had previously been accepted as the " Brown-tail " or " Cul brun ", really represents the " Gold-tail " or " Cul doré ". Although this opinion was discussed and agreed to by such writers as Barnes and Benjamin in 1924 (Proc. ent. Soc. Wash., **26**, p. 213) and Joannis in 1928 (Encyc. ent., Sér. B III, Lep. **3**, p. 99), the great majority of systematic works since published in Europe still adhere to the interpretation of *P. chrysorrhoea* as the Brown-tail.

In 1936, when arranging the collection of Lymantriidae in the British Museum (Nat. Hist.), I investigated the question as fully as possible, and decided to retain *P. chrysorrhoea* as the Brown-tail, but as the matter is still the subject of discussion, my reasons for this course are set out below.

Linnaeus' description in Syst. Nat. 1758, X, 1, p. 502, is as follows :—

> Chrysorrhoea. 28. *P. Bombyx* elinguis, alis deflexis albidis, abdominalis apice barbato luteo.
>
> *Raj. ins.* 156. *n.* 1 : 15. *Reaum. ins.* 1. *phal.* 2.*t*.22.
> *Merian. eur.* 1.*p*.8.*t*.20. *Roes. ins.* 1. *phal.* 2.*t*.22.
> *Frisch. ins.* 3.*p*.28.*t*.8. *Wilk. pap.* 28.*t*.3.*b*.3.
> *Edw. av.* 122.*t*.122.
>
> *Habitat in* Pomonae *arboribus*, Quercu, Alno.
> Larva *nodosa, pilosa, nigra, rubro lineata.*

Rothschild argued that " barbato luteo " represents the Gold-tail, and that Linnaeus' first quotation, of Ray, states that the larva has " three scarlet lines, one situated above the feet on each side of the body ", whereas the Brown-tail has no lateral lines. He also says that Linnaeus' own mention of the larva, with " rubro lineata " also proves that the Gold-tail was intended.

Ray's statement regarding the *lateral* line on the body is the only one which seems to me conclusively to represent the Gold-tail—the larva of the Brown-tail has two red lines dorsally. As regards the " barbato luteo ", luteo meaning golden yellow, saffron yellow or orange yellow, any series of the two species will show specimens with the same shade of deep orange on the anal tuft, and occasional examples in which the " yellow " and " brown " are reversed. Nordström and Wahlgren's Svenska Fjärilar, commenced in 1935, dealing with the Lepidoptera of Linnaeus' own country, shows on plate 14 a picture of each species with the anal tuft of the same shade of orange, and incidentally cites *P. chrysorrhoea* as the Brown-tail.

An examination of the original collection of Linnaeus in the possession of the Linnaean Society of London shows 1 ♂ 4 ♀♀ Gold-tails and 1 ♂ 3 ♀♀ Brown-tails. On an unset ♀ specimen of the Brown-tail is a label *in the handwriting of Linnaeus*, " *chrysorrhoea* ". A pair of Gold-tails bear labels " *auriflua* Fab." in the handwriting of Smith, who formerly owned the collection, and the remaining specimens are unnamed. It should be mentioned that the abdomen of the specimen labelled " *chrysorrhoea* " has never been detached, and the transposition of the label is the only (and a most unlikely) possibility.

The two species are uncommon, or even rare, in Sweden, and the seven references given by Linnaeus, with one exception (that of Roesel), deal with only one insect. It follows that Linnaeus may not have realised in 1758 that two species, admittedly similar, were in question. The fact that vein M3 is absent in the hindwing of the Gold-tail was missed by the early authors.

The references given by Linnaeus do not agree in their description of the insect, and seem to show that either he considered his *P. chrysorrhoea* to be very variable, or that he did not trouble to study the details carefully. He arranged the references somewhat in order of date.

The salient points of the references are as follows :—

Raj. ins. 156.n.1 : 15. (John Ray, Historia Insectorum, 1710). As Rothschild states, the description of the moth might fit either of the two insects, but the larva is the Gold-tail.

Merian. eur. 1,p.8.t.20. (Merian, Histoire des Insectes de l'Europe, 1730). Describes a white nocturnal moth, illustrated by a poor and uncoloured representation of larva and perfect insect. The species intended is uncertain.

Frisch. ins. 3. p.28, t.8. This reference appears to indicate Frisch's Insecten in Teutschland, 1730. This was reprinted later with slight alterations under the date of 1766. The reference cannot be traced, Part III, plate II, fig. 1 gives an uncoloured figure of larva and both sexes of an obvious Gold- or Brown-tail, but which of these is uncertain.

Edw. av. 122.t.122. (George Edwards. A Natural History of Birds, 1750). A rather poor illustration of a moth, in colour. In the text is a reference to the "orange colour at the tip of the tail". The species intended is uncertain, the Gold-tail the more probable.

Reaum. ins. 1. phal. 2.t.22. This reference cannot be traced, but is perhaps to Reaumur's Mémoires pour servir à l'Histoire des Insectes, 1734, Tom. 1, pl. 2, page 108, mem. 2. On this plate there are 21 figures only, but the "21" is badly engraved and resembles "22". The figure is a larva, apparently *Dasychira fascelina* Linnaeus. In this same volume, Tom. 1, pl. 16, page 342, mem. 7, figs. 8–11, represent a Gold- or Brown-tail, the explanation of the figures being on page 332. Identity is uncertain, but perhaps the Brown-tail. An edition bearing the date 1737 has the same plates but different page numbers.

Roes. ins. 1. phal. 2.t.22. (Roesel's Der Monatlich herausgegeben Insecten Belustigen, 1746–61). An excellent coloured plate and description of larva and both sexes of the perfect insect, readily identifiable as the Brown-tail. Tab. 21 is an equally good picture of the Gold-tail.

Wilk. pap.28.tab.3.b.3. (Wilkes' English Butterflies and Moths, 1747–60). An excellent coloured plate and description of larva and both sexes of the Gold-tail. Wilkes also gives a reference to Roesel tab. 21. The Brown-tail is not mentioned.

To recapitulate these seven references, we have four uncertain, two to the Gold-tail (Ray and Wilkes), and one to the Brown-tail (Roesel). Roesel's, however, is the only case in which the two species are shown and the differences brought out.

The first author to write in detail after the publication of Linnaeus' Xth edition was apparently Scopoli, Entomologia Carniolica, 1763. Under *Phalaena chrysorrhoea*, on page 197, fig. 493, he mentions the anal tuft as "flavo", but gives two references, the first to Linnaeus' Syst. Nat. p. 502, the second to Roesel's "Papil. Noct. Cl. II, Tab. 22, fig. 4.5." The latter appears to be a shortened reference to the book cited above, but the same plates and numbers are used by Roesel in different editions. Scopoli therefore selects the Brown-tail, but does not mention the Gold-tail.

Linnaeus' XIIth edition of 1766, page 822, gives the same seven references as in the Xth edition, but with two additions. One of these, placed first in the list, is

" Scop. Carn. 493 " referred to above. The other, placed near the end, is Geoffr. Paris, 117.n.20 " (Histoire abregée des Insectes . . . de Paris, Geoffroy, 1762), in which the insect is styled " La phalée blanche a cul brun ".

Fuessly, 1775, Schweitzerischen Insecten, 35, names the Gold-tail as *Phalaena similis* Fuessly, and cites Roesel's Tab. 21 and 22 for this species and *P. chrysorrhoea* respectively.

To sum up, it appears to me that in view of the named specimen in the collection of Linnaeus, his selection of Roesel's plate 22, rather than plate 21, in the only reference in which the differences between the two insects are brought out, and his selection of Scopoli as his first reference in the XIIth edition of his Syst. Nat., clearly show that the weight of opinion among entomologists for the Brown-tail or Cul Brun is justified, and should not be reversed.

As regards the genus the earliest name is *Euproctis* Hübner, 1819, Verz. bekannt. Schmett., **10,** p. 159, type fixed by Moore, 1882, Lep. Ceylon, **2,** p. 89 as *Phalaena chrysorrhoea* Linnaeus 1758.

Of other genera which have been used by some authors, *Arctornis* Germar, 1811, type fixed by Kirby, 1892, Cat. Lep. Het., **1,** p. 432 as *Phalaena l-nigrum* Müller 1764, is not congeneric with *P. chrysorrhoea* Linnaeus. *Nygmia* Hübner, 1820, type fixed by Kirby, 1892, Cat. Lep. Het., **1,** p. 448 as *Nygmia icilia* Stoll, 1790, is later than *Euproctis*.

The name to be applied to the Gold-tail or Cul doré should be *Phalaena similis* Fuessly, 1775, which takes precedence over *Bombyx auriflora* Denis and Schiffermüller 1775 on the evidence set out by Prout in 1900 in Ann. Mag. nat. Hist., (7) **6,** p. 158.

Many systematists place *Euproctis chrysorrhoea* Linnaeus and *Euproctis similis* Fuessly in different genera, but a study of African and Oriental species appears to me to discourage this view.

I am much indebted to Mr. W. H. T. Tams for assistance in the writing of this paper.

LABORATORY STUDIES ON THE BIONOMICS OF THE RAT FLEAS, *XENOPSYLLA BRASILIENSIS*, BAKER, AND *X. CHEOPIS*, ROTHS.

II. Water Relations during the Cocoon Period.

By E. B. Edney, Ph.D.
*Tutor in Biology, Makerere College, Uganda.**

Introduction.

In Part I of these studies (Edney 1945) it was shown that longevity of unfed adult fleas bears a linear relationship to the saturation deficit of the air to which the cocoon stage is exposed. In view of Mellanby's (1933) statement that the true pupae are water savers, the suggestion was put forward that, during the short period between cocoon formation and pupation, the insects lose water in relation to the saturation deficit of the air, and finally emerge as adults with a greater or smaller reserve of water depending on the saturation deficit they have experienced during the short critical period.

The work to be described was undertaken with a view to confirming or disproving the above suggestion and to obtaining more precise data upon the water relations during the whole of the cocoon period and during the first few days in particular.

The sequence of events from the time the larva ceases to feed until the adult flea emerges has been thoroughly worked out by Sharif (1937), whose observations have been repeatedly confirmed by the writer. The relevant points may be summarised as follows.

The third-stage larva, after a variable period, stops feeding, expels all the contents from the gut, and within 24 hours spins a silken cocoon which becomes covered outside with particles of débris. Then follows a resting period of about three and a half days, during which the larva lies bent double inside the cocoon. There has been some doubt as to the correct terminology of this period ; in the present paper the term ' prepupa ' will be given to the insect and ' prepupal period ' will be used for the time between final defaecation and the next moult. The larva, then, after resting for a few days, undergoes ecdysis into a white exarate pupa. After about four days the white cuticle darkens and after a further period of variable length the adult emerges from the pupal skin and from the cocoon.

Experiments.

Fleas were bred on baby white mice in jars, using a technique similar to that of Leeson (1932). All the fleas used in the experiments to be described were *Xenopsylla brasiliensis*, Baker.

In order to get a general impression of what happened during the cocoon period, it was decided as a preliminary experiment to weigh batches of insects, together with their cocoons, daily throughout the whole period. Further experiments could then be devised in the light of information obtained.

1. *Variation in weight throughout the cocoon period.*

Batches of from fifty to a hundred freshly formed cocoons containing prepupae were obtained from the culture jars each day. Each batch was weighed and placed in a squat pattern weighing bottle which was kept in a desiccator where the humidity was controlled by KOH solution. The relative humidities used were 90, 80, 70, 60, 50, 40, 30 and 20 per cent. On one day the freshly formed prepupae were separated from their cocoons, and cocoons and prepupae kept as separate batches. Another control batch consisted of bedding material from the culture jar, including sand, a little bran and a small amount of dried blood. These last three batches were all kept at 70 per cent. R.H.

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* Now Lecturer in Zoology, University of Birmingham.

All the desiccators were kept together in the dark in a room where the temperature could be controlled within fairly narrow limits. The precise temperature conditions in this room have already been described in Part I of these studies, and since the conditions during the experiments to be described did not differ in any important respect, the mean temperature, 24°C., will again be used to represent temperature in the dark room.

Each batch was weighed once a day from the day of cocoon formation until all the adults had emerged. At each weighing, in order to reduce to a minimum the exposure of cocoons and their contents to room air, the lid of the desiccator was removed with as little disturbance as possible and a close fitting lid was placed on the weighing bottle containing the cocoons before removing it from the desiccator. During the period when adult fleas were emerging, their weight was allowed for in subsequent days weighings.

The results are shown in fig. 1. In order to compare the variation in weight of different batches the weight of each has been expressed as for 100 insects, the actual number used in each batch being stated in the legend to the figure.

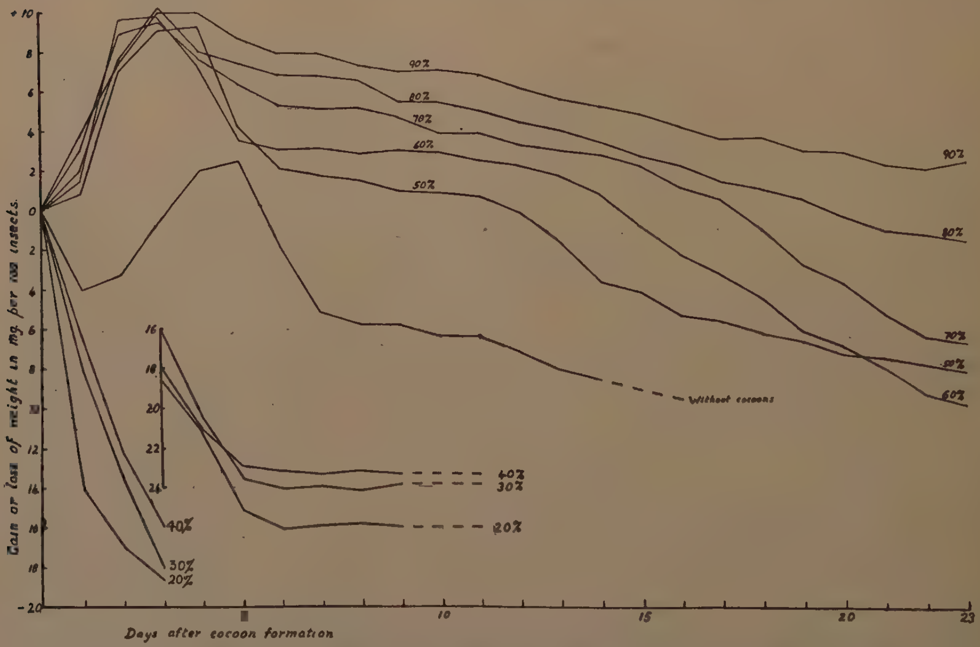

Fig. 1.—The variation in weight of batches of insects and their cocoons, in relative humidities from 20 to 90 per cent., during the cocoon period. Variation in weight is expressed in mg. per hundred insects ; the actual number used in each batch being as follows :—20 per cent., 64 ; 30 per cent., 45 ; 40 per cent., 68 ; 50 per cent., 83 ; 60 per cent., 56 ; 70 per cent., 50 ; 80 per cent., 61 ; 90 per cent., 48.

The empty cocoons and the bedding did not change in weight significantly once they had come into equilibrium with the humidity (70 per cent.) in which they were kept and they have not been graphed in fig. 1. The other curves show several interesting features, the most striking of which is the sharp difference in behaviour between those insects kept at 50 per cent. R.H. or above, and those kept at a lower humidity. The former, instead of losing weight as might be expected, show a rapid increase in weight for the first three days. The latter, on the other hand, begin to lose weight rapidly and continue to do so more and more slowly for some time. Examination of these low humidity batches at the end of the experiment showed that they had all shrivelled up and only one insect (in 40 per cent. R.H.) had pupated. Furthermore, the amount by which those batches in 50 per cent. or above gain weight does not appear to depend upon the humidity, for they all gain more or less equally.

After the first three or four days, those insects which have gained weight begin to lose it, and continue to do so until they emerge, the curves here suggesting that the rate of loss of weight is determined by the humidity of the air.

The amount of weight gained during the first three days under the conditions of the present experiment varies from 9 to 10 mg. per hundred prepupae. If this increase is ascribed to the insects alone, it amounts to an increase of 26–29 per cent. since one hundred freshly formed prepupae weigh approximately 35 mg. It would, however, be unwise without further evidence to assume that all the weight is gained by the insects, for the control cocoons themselves gained a small amount, and the association between cocoon and contained prepupa might well behave in a very different way from the sum of their separate behaviours.

The behaviour of the prepupae which had been removed from their cocoons is interesting. During the first day they lost weight, but in the following four days they gained more than 6 mg. per hundred, and then began to lose again. It was noticed that after the first day these prepupae had spun new rough cocoons of silk, and since their weight subsequently began to rise it seemed reasonable to ask whether a covering of silk was in some way necessary for an increase in weight. That it is not necessary, though it may encourage the process, was shown by a further experiment to be described later.

No reliable quantitative information can be obtained from the preliminary method of approach described above, for it is open to several sources of error ; for instance, it is not possible to say whether any of the insects have died inside their cocoons until the end of the experiment, and even then it is not possible to say when they died. Insects which died would, of course, lose weight, and might affect the results very considerably. Another source of error lies in the fact that the weights of insect and cocoon are recorded together, and there is no accurate measure of the insect's weight by itself.

The whole experiment was repeated, and the results did not differ in any important respect from those already described, except that the initial gain in weight was somewhat lower (from 6 to 8 mg. per 100 insects). The results obtained suggest a number of questions. At what stage in their development do the insects cease gaining weight and begin to lose it ? Is the gain in weight due to absorption of water from the air ? Is the amount gained affected by the presence of cocoon material round the prepupae ? Is the gain really independent of humidity of the air above a certain point ? If so, what is the limit of humidity below which insects dry out ? Does this lower limit vary with temperature ? After the initial gain in weight, is the subsequent fall due to loss of water and does it vary with the humidity ? The experiments which are described below were carried out in an attempt to answer these questions.

2. *The stage in development at which gain in weight ceases and loss begins.*

It is clear from fig. 1 that most of the insects increase in weight up to three days from the commencement of the prepupal period and begin to lose it during the fourth day, with the exception of those in 90 and 50 per cent. R.H. which remained constant during the fourth day in the experiments shown in fig. 1, but lost weight during the same day when the experiment was repeated. The prepupal period is known to last about three or four days at 24°C., and it therefore appears likely that the change from gain to loss takes place when the prepupa moults into the pupa. Several series of cocoons which had been kept at various humidities were opened after three days and other series were opened after four days. At all relative humidities from 50 to 90 per cent. the vast majority had pupated after four days and very few after three days. It can, therefore, be said that at all humidities in which prepupae gain weight the change from gain to loss begins about the time of moulting.

3. *The water content of prepupae before and after increase in weight.*

If the gain in weight observed is due to the absorption of water, then the percentage of water in prepupae should be higher at the end of the three day period during which weight is gained than at the beginning.

Freshly formed cocoons were obtained each day and divided into two approximately equal batches. The first batch was kept in a squat pattern weighing bottle in a desiccator at a constant relative humidity of 80 per cent. The second batch was used for determining the initial weight and water content. For this purpose the prepupae were caused to emerge from their cocoons, which they readily do upon being agitated or under slight pressure. Any which showed the slightest sign of being damaged were discarded. They were then weighed and dried in air at 100°C. to constant weight.

After three days, the prepupae of the first batch, which had been left undisturbed, were removed from the desiccator and dissected out of their cocoons. The process of dissection can, with a little practice, be performed quite rapidly and without damaging the turgid insects. As each prepupa was dissected out it was transferred by a fine brush to a small closed weighing bottle. It was thought that loss of water during the time between dissection and weighing might lead to error, but any such error was shown to be insignificant by weighing two batches of prepupae immediately after dissection, keeping them for one hour in a small closed weighing bottle in laboratory conditions, and weighing them again. No significant difference in weight was found.

After dissection the prepupae were weighed and then dried to constant weight. By this means it was possible to estimate the change in weight during the three day period and also the change in water content.

Five batches, obtained on successive days, were used, and the results are summarised in Table I. It is clear from these figures that the percentage increase in weight during the prepupal period varies considerably (from 10 to 20 per cent. in the five batches used). It is also clear that the percentage of water present in the insects increases during the same period, though here again the amount by which it increases varies from one batch to another.

TABLE I.

The weight and water content of prepupae less than 24 hours after cocoon formation, compared with the same measurements after being kept for three days at 80 per cent. R.H. and 24°C. Small numbers in brackets refer to the number of insects used for each measurement.

Initial mean weight × 100 (in mg.)	Initial water content (percent. of wet weight)	Mean weight after three days × 100 (in mg.)	Water content after three days (percent. of wet weight).	Increase in weight (percent. of original weight).
34·5 (37)	64	41·5 (40)	74	20
38·3 (58)	65	43·5 (54)	70	13
34·3 (84)	67	37·7 (76)	72	10
36·6 (105)	65	42·9 (90)	75	17
36·6 (86)	65	40·5 (72)	73	11
Mean	65·2	—	72·8	14

Working with the mean figures obtained from the five series we may now argue as follows : initially, 100 g. of prepupae contain 65·2 g. water and 34·8 g. dry material, and each 100 g. of prepupae increases in three days to 114 g., of which 72·8 per cent., or 83 g. is water, and 31 g. is dry matter. It follows that 3·8 g. of dry matter have been lost and 17·8 g. of water gained. The insects do not defaecate during the prepupal period, so the loss of dry material must be due to metabolism of food reserves. Fat is frequently used by insects during resting periods (Wigglesworth 1930, p. 329) and the complete oxidation of fat to CO_2 and water yields 107 g. water for every 100 g. fat (Shohl 1939). Therefore, if the whole of the dry material lost is fat and is completely oxidised, and if the water of oxidation is retained by the insects, it would account for nearly 4·1 g. of water, leaving the difference between this and the total amount of water gained, 13·7 g., to be accounted for by absorption from the air. If the dry material lost is not fat, or is not completely oxidised or if the resulting water is not all retained, then so much more water must have been absorbed from the air.

It can be said then that a minimum of 13·7 and a maximum of 17·8 g. of water are absorbed per 100 g. of prepupae, and an assumption that the amount of water absorbed is equal to the total weight gained would not be far wrong.

4. *The effect of the absence of cocoon material on the weight gained by prepupae.*

Three series of prepupae obtained on three successive days were used in this experiment. In the first series the prepupae were caused to emerge from their cocoons and weighed; they were then put back with their cocoons and most of them re-entered. The second series was treated in the same way as the first except that after weighing the insects were deprived of their original cocoons. In the third series, the prepupae, after being weighed, were put in a narrow-necked bottle which was held horizontally and rotated by clockwork, and this agitation was sufficient to prevent their spinning more silk. The bottle was left unstoppered and was kept, together with the clockwork mechanism, in a large airtight trough over KOH maintaining a relative humidity of 80 per cent. The first two series were also placed in similar bottles at the same humidity, but these were not rotated. After two days all the prepupae in series 3 were seen to have folded and become motionless, and only a trace of silk was present. The rotating mechanism was therefore stopped. After three days the prepupae were weighed again, in series 1 and 2 after dissection from their cocoons. At 80 per cent. R.H. and 20°C. 105 prepupae of series 1 gained 24 per cent. of their original weight, 67 of series 2, 10 per cent., and 79 of series 3, 8 per cent. From these results it is clear that all series increased in weight, though the prepupae without their original cocoons gained less than those with, and the prepupae prevented from spinning silk gained least of all.

It can be concluded from these results that silk and cocoon material are not necessary for the prepupae to gain in weight although their presence allows a greater increase. But it has not been shown that the greater increase is due to any physiological relation between cocoon and insect, for the smaller amount gained in series 2 may well be explained by abnormal loss of water in spinning fresh silk, and in series 3 by increased metabolism as the result of agitation and movement.

5. *The effect of prepupae on the humidity of the air in a small enclosed space.*

The absorption of water vapour from the air by prepupae during the first three days was demonstrated by the following experiment. A batch of fifty freshly formed cocoons with their contained prepupae was placed together with a small " Edney " paper hygrometer on an inverted Petri dish and covered with another slightly smaller dish. The edge of the latter was sealed with wax. A similar apparatus, but without cocoons or insects, was set up as a control, and both were kept in a dark room at about 24°C. The two hygrometers were previously adjusted to read correctly at

70 per cent. R.H., or roughly the humidity in the room when the experiment was started.

The two hygrometers were read every few hours during the first three days of the experiment, and then once or twice every day until adult fleas began to emerge. Any change in the humidity of the air enclosing the hygrometers due to small variations in room temperature was revealed by the control hygrometer, and in recording the variations of the other hygrometer these deviations were allowed for. After making this adjustment the humidity in the dish enclosing the insects has been graphed in fig. 2. The curve falls very rapidly during the first day, until the R.H. has fallen to about 50 per cent. It varies very little (within a range of 2 per cent.) during the next 48 hours and then rises very rapidly, until, 5 days from the start, it has returned to a little above the original figure, where it remains nearly constant, but rising slowly, for the next five or six days. After this the rate of rise very gradually increases until the seventeenth day, when the adults begin to emerge.

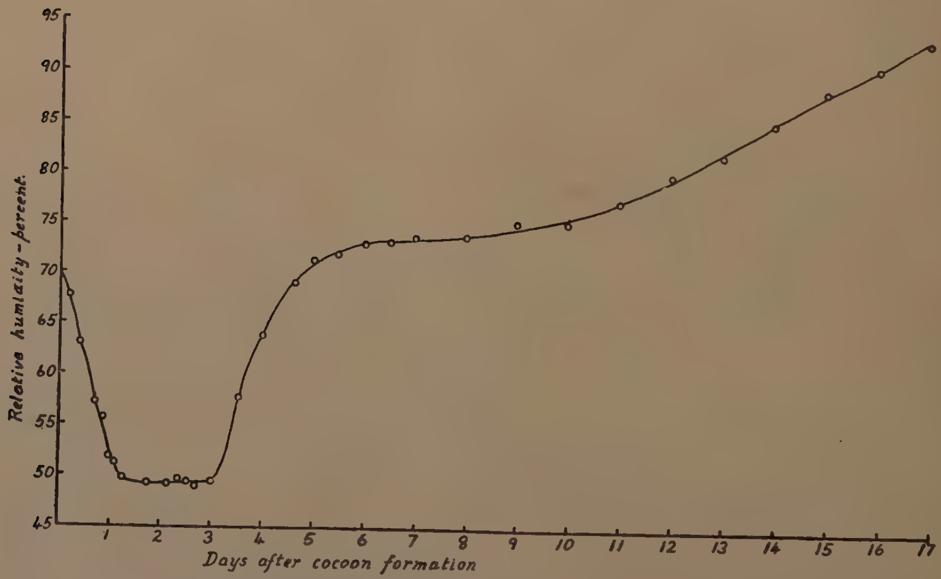

Fig. 2.—The variation in relative humidity in a closed vessel containing about 90 ml. of air and 50 insects and cocoons, during the cocoon period.

The hygrometers used are not accurate over a wide range, but by re-calibrating the instrument at the end of the experiment it was possible to estimate fairly accurately that the lowest humidity to which the air had fallen was 47 per cent. R.H. The re-calibration was done by subjecting the instrument to 70 per cent. R.H. again and then finding the readings at known humidities of 45 and 50 per cent.

In order to interpret the curve, which resembles in many respects the reverse of the weight curve in fig. 1, the actual quantities of water involved must be considered. From fig. 1 we know that 100 prepupae in their cocoons gain approximately 10 mg. during the first three days, and a batch of 50 could therefore be expected to gain 5 mg. Now the volume of air inside the sealed Petri dish in the present experiment was roughly 90 ml., and this volume of air at 24°C. contains 1·36 mg. water vapour at 70 per cent. and 0·91 mg. at 47 per cent. R.H. The insects had therefore only to absorb 0·45 mg. of water to reduce the R.H. to 47 per cent. and they appear to have done this in the first 24 hours. But fig. 1 also shows that in 40 per cent. R.H. the insects are incapable of absorbing water, and it seems likely, therefore, that the prepupae after having reduced the R.H. to 47 per cent. were incapable of absorbing more water from so dry an atmosphere and came into equilibrium with it at that humidity. This state persisted for two days, after which, it is suggested, the prepupae moulted, and immediately began to lose water rapidly, as did those in low humidities shown in fig. 1. From this point onwards the curve corresponds in reverse with the weight curves in fig. 1, and need not be discussed further, except to remark that the end of the curve, which appears to rise as far as 95 per cent. R.H., is probably too high, since paper hygrometers cannot be relied upon at high humidities.

6. *The effect of humidity upon absorption of water during the prepupal period.*

The curves in fig. 1 do not indicate any close relation between humidity and the amount of weight gained during the prepupal period, and the present experiment was designed to obtain further information on this point. Prepupae, obtained in the usual way, were caused to emerge from their cocoons, weighed and returned to their cocoons. Each day's batch was then kept in one of the relative humidities 60, 70, 80, or 90 per cent. at 24°C. for three days, after which the prepupae were dissected out and weighed again. Any prepupae which had not returned into a cocoon after weighing on the first day, or which had pupated, were discarded.

TABLE II.

Increase in weight of prepupae at various humidities during the prepupal period. The small numbers in brackets refer to the numbers of insects used in each batch.

Relative humidity percent.	Initial mean weight × 100 (in mg.)	Mean weight × 100 after three days (in mg.)	Increase in weight (percentage of initial weight)
60	49·1 (33)	52·4 (30)	7
60	38·1 (53)	46·2 (45)	21
70	46·3 (52)	52·4 (43)	13
70	37·2 (52)	45·8 (44)	23
80	37·2 (36)	43·5 (30)	17
80	43·0 (51)	50·0 (47)	16
80	37·3 (43)	46·5 (35)	25
90	42·2 (28)	48·7 (27)	15
90	38·1 (53)	44·0 (47)	15
90	38·2 (40)	43·8 (36)	15

The results are shown in Table II. Here again the increase in weight shown by various batches varies considerably. But there is no indication of a relation between humidity and amount of weight gained.

7. *The effect of humidity during the prepupal and pupal periods upon change in weight and water content and upon adult longevity.*

From the results of the previous experiments it is clear that each day's batch of prepupae varies both in weight and in subsequent behaviour from other batches, so that any critical investigation of the effect of humidity must be made by subjecting parts of the same batch of prepupae to different conditions. The disadvantage of this procedure is that it results in having small numbers of insects in each batch, but this can be largely overcome by repeating the experiment several times, by reducing the number of different conditions, and by breeding very large numbers of fleas.

For this experiment, therefore, it was decided to use three humidities only, 50, 70 and 90 per cent. R.H., and the experiment was carried out at a time when the breeding jars were producing large numbers of prepupae each day. Rather more than a hundred cocoons were obtained from each of two breeding jars each day for a period of six days, and they were used as follows. On each day, all the prepupae from the first jar were collected and given an odd serial number (1 on the first day, 3 on the second and so on), while the cocoons from the second jar were given an even serial number. Every day a sample of about 30 was taken from each series and weighed, and the water content of the odd and even series was found on alternate days. The remainder in each series was divided into three batches labelled A, B and C, and in both odd and even series, on those days when only the weight and not the water content had been estimated, the insects which had been weighed were returned in equal numbers to the three batches. All the A batches were kept at 50 per cent., all the B's at 70 per cent. and all the C's at 90 per cent. R.H. Those batches which had even serial numbers were removed after three days, weighed again and their water content found, while the batches with odd serial numbers were left until they emerged as adults, when they were weighed and each insect was put into a separate tube and kept to find the longevity at 80 per cent. R.H. Each batch of insects was kept in a squat pattern weighing bottle, covered towards the end of the pupal period with silk voile in the odd numbered series to prevent the emerging adults from escaping.

It was originally planned to find the water content of the adults emerging from each humidity in three of the odd numbered series, and to use the other three for longevity trials ; but in the event it was found that with one exception only a small number of adults emerged from any batch on one day, and that, because of this, measurement of the water content would be liable to too great an error. It was decided, therefore, to estimate the water content of adults from the different humidities in another experiment except on one day when a sufficiently large number of adults emerged from one series (No. 5) to make it worth while estimating their water content.

By following the procedure outlined above, the six batches of insects used on each day formed by themselves a self-contained experiment, though with somewhat small numbers, and the whole was repeated six times. The results are summarised in Table III, and a few remarks must now be made on the methods used in arriving at the figures there set down.

The weights are, as usual, expressed as the mean weight × 100 in mg., and small figures in brackets show the number of insects on which each mean is based. Water content is expressed as a percentage of the total wet weight.

Newly emerged males are a little lighter than females, so that the mean weight of a batch of adults which emerged on any one day cannot be used for comparison with other batches unless each consists of males and females in equal proportions. In order to obtain a representative figure, the ratio of male to female weight was calculated after weighing three groups of freshly emerged insects (not used in the present experiment) with the following results :—

In one group, 76 males weighed 22·2 mg. (average weight 292 units)
and 68 females ,, 27·2 mg. (,, ,, 400 ,,)
in another group, 92 males ,, 27·0 mg. (,, ,, 293 ,,)
and 78 females ,, 30·0 mg. (,, ,, 385 ,,)
in a third group, 70 males ,, 21·4 mg. (,, ,, 305 ,,)
and 84 females ,, 32·8 mg. (,, ,, 390 ,,)

If the mean weight of males is expressed as a percentage of the mean weight of females in each case, the figures are 73, 76 and 78 per cent. respectively, and the ratio of 3 to 4 was used as being at once convenient and very near to the middle of the range of variation found. On two occasions, as a confirmation, the adults being used in the present experiment were themselves divided into males and females and weighed separately, and the ratio of 3 : 4 was very closely approximated.

The number of males and females in each batch was found after they had all died and a correction was applied based upon the ratio mentioned, as follows : if a batch of adults which gave a mean weight of 330 units per flea was found to consist of 15 males and 5 females, a simple calculation shows that a male weighs 305 units and a female 407. A weight half way between these two, that is, 356 units, was then taken to represent the mean weight of the batch, and this has been called the corrected weight. If multiplied by a hundred it represents the corrected mean weight of an insect × 100 in mg.

It may be objected that since adult males and females are of different weights it is possible that the two sexes differ in weight in the prepupal stage also, and that if so, a correction should be applied there as well. However, the total number of males which emerged in any batch was very nearly equal to the total number of females (it was only on separate days that the proportions varied), so that the sexes were equally represented in the prepupal stage and a correction becomes unnecessary.

The length of life is expressed in days. The fleas were examined only once a day, so that an insect living for, say, ten and a half days would be recorded as having lived for eleven days.

In each batch there were usually two and sometimes three " emergence groups ". The first group consisted mainly of females, and the last mainly of males (it is well known that the female pupal period is shorter than that of the male), and the intermediate group was a mixture of the sexes. Experiments not recorded in the present paper suggest that the longevity of newly emerged, unfed males, is slightly greater than that of females, but, even if the difference proves to be real, it is very small and therefore the mean longevity of all adults in each batch was used to represent the longevity of that batch. It is perhaps worth recording that the longevity figures were also calculated by considering each sex separately, and they showed the same general tendency, that is, for adults to live longer if their cocoon period has been passed in a high humidity ; except for the males of series 11, where the figure in column C was lower than that in column B by 0·5 days.

The results set out in Table III may now be considered in more detail. Considering first the weight at the beginning and end of the three day prepupal period, we find that an increase was shown in all batches, no matter at what humidity they were kept, and further that there is no clear indication that humidity has any effect upon the amount of weight gained. If columns A and B alone are considered there is a suggestion that those prepupae in 70 per cent. R.H. gained slightly more than those in 50 per cent., for the figure in column B is either equal to or slightly greater than the corresponding figure in column A in each series, but the significance of the difference is highly doubtful.

TABLE III.

The effect of humidity during the cocoon period on weight and water content of prepupae after three days, and on weight and longevity of adults. Figures in brackets refer to the number of insects used for each measurement.

Date of cocoon formation	Serial number	Initial mean weight of prepupae × 100 (in mg.)	Initial water content (percentage of wet weight)	Mean weight of prepupae × 100 after three days in various humidities (in mg.)			Water content of prepupae after three days in various humidities (percentage of wet weight)			Corrected mean weight of adults × 100 (in mg.)			Longevity of adults (days)				
				A	B	C	A	B	C	A	B	C	A	P	B	P	C
26·2	1	33·0 (20)	70	—	—	—	—	—	—	24·5 (25)	27·5 (26)	30·5 (32)	15·2	0·4	17·7	< 0·01	21·5
26·2	2	36·0 (30)	—	40·0 (25)	41·0 (24)	38·0 (25)	70	72	75	—	—	—	—	—	—	—	—
27·2	3	29·0 (30)	—	—	—	—	—	—	—	24·5 (33)	26·5 (30)	24·0 (39)	16·4	0·42	17·4	0·76	17·7
27·2	4	31·0 (30)	72	37·5 (27)	40·0 (21)	39·0 (28)	76	75	75	—	—	—	—	—	—	—	—
28·2	5	29·3 (30)	68	—	—	—	—	—	—	20·5 (53)	22·0 (50)	24·0 (27)	—	—	—	—	—
28·2	6	31·0 (20)	—	34·5 (32)	34·5 (36)	35·5 (33)	77	79	77	—	—	—	—	—	—	—	—
1·3	7	20·7 (30)	—	—	—	—	—	—	—	17·0 (43)	20·0 (61)	22·0 (41)	9·5	< 0·01	11·8	< 0·01	13·5
1·3	8	26·0 (30)	67	29·5 (26)	31·0 (28)	32·0 (32)	74	72	75	—	—	—	—	—	—	—	—
2·3	9	21·3 (30)	72	—	—	—	—	—	—	15·0 (26)	16·0 (37)	17·0 (33)	11·3	0·48	11·8	0·55	12·4
2·3	10	27·0 (20)	—	28·0 (30)	29·0 (34)	28·0 (34)	75	76	74	—	—	—	—	—	—	—	—
3·3	11	24·0 (30)	—	—	—	—	—	—	—	17·5 (18)	18·5 (29)	19·0 (27)	11·5	0·65	12·0	0·61	12·5
3·3	12	19·0 (20)	70	20·5 (27)	21·0 (26)	21·5 (28)	76	78	70	—	—	—	—	—	—	—	—
		Mean :	69·8				74·7	75·3	74·3	Combined probability : P < 0·01						< 0·01	

The letters A, B and C refer to relative humidities during the cocoon period of 50 per cent., 70 per cent. and 90 per cent. respectively.

If the increase in weight shown by each batch is expressed as a percentage of the initial weight of the prepupae, the following figures are arrived at :—

Series No. ...	2	4	6	8	10	12	Mean
Relative humidity 50 per cent.	11	21	11	13	4	8	11·3
,, ,, 70 ,, ...	14	29	11	19	7	10	15·0
,, ,, 90 ,, ...	6	26	14	24	4	13	14·5

The variation is large, particularly the variation between one series and another, which is in fact greater than that between the various humidities within a series. Thus series 10 shows all low percentage increases (4, 7 and 4), while series 4 shows high ones (21, 29, and 26) over the same range of humidities. The means, for what they are worth, do not suggest a relation between humidity and increase in weight.

Turning now to the water content figures at the beginning and end of the prepupal period, we find that here again there is a significant increase during the three days, but that this increase does not bear any obvious relation to the humidity. The mean percentage water content in prepupae less than 24 hours old is 69·8, and the mean percentages after three days in 50, 70 and 90 per cent. R.H. are 74·7, 75·3 and 74·3 respectively, and the differences between these figures are certainly not significant.

The loss in dry weight per 100 g. of prepupae was 1·7, 1·9 and 0·9 g. in 50, 70 and 90 per cent R.H. respectively, and assuming that these losses all represent completely oxidised fat, the minimum amounts of water absorbed from the air are 12·9, 14·9 and 14·4 g. per 100 g. in the three humidities respectively.

As regards the weights of adult insects, the figures do suggest a relation to humidity. In each series (except No. 3) the adults in column C are heavier than those in column B, and the latter are in each series heavier than those in column A.

The longevity of the adults shows a corresponding variation. In all five series tested, adults from 90 per cent. R.H. lived longer than those from 70 per cent. and the latter lived longer than those from 50 per cent. Statistical analysis of the difference between A and B and between B and C in each series, shows that in three series both differences are not significant (P is greater than 0·05) whereas in the other two series both differences are significant. In a series of parallel experiments such as this, it is possible to combine the probabilities derived from individual comparisons and to obtain an aggregate probability of significance. This has been done according to the method described by Fisher (1941) section 2·11, and the difference is found to be highly significant (P is less than 0·01) in each case.

In only one series was the water content of the adults determined, and the results, though in conformity with the rest of the figures, are not by themselves conclusive (they were 74, 76 and 77 per cent. for groups A, B and C respectively) and a separate experiment was made to find the effect of the three humidities during the pupal period upon the water content of adults.

8. *The effect of humidity during the pupal period on the water content of newly emerged adults.*

Since it has already been shown that the humidity has no obvious effect during the prepupal period, provided it is above 50 per cent. R.H., the prepupae in this experiment were all kept at 80 per cent. R.H. and put into different humidities after four days, when they could be assumed to have moulted into true pupae. Six batches of prepupae were used, and for the pupal period each was divided into two, the halves being kept at either 50 and 70, 70 and 90, or 50 and 90 per cent. R.H. On the nineteenth day after cocoon formation (fifteenth after pupation), the adults were recovered.

Most of the adults emerged upon agitation, the rest were caused to emerge by gently pressing the cocoon, and in this way it was possible to obtain nearly all the adults from each batch for weighing in one emergence group. The few which did not emerge were discarded.

In expressing the results (Table IV), no correction was applied to the mean weight of each batch of adults because the proportion of males to females, though varying somewhat from series to series, was approximately the same in each half of every series.

TABLE IV.

The effect of humidity during the pupal period on adult weight and water content. Small figures in brackets refer to the number of insects used for each measurement. Prepupae and pupae kept at 24°C.

Series	Relative humidity percent. during the pupal period	Mean weight of adults × 100 (in mg.)	Water content of adults (percent. of wet weight)
1	90	34·3 (32)	68·8
	70	32·5 (32)	65·4
2	70	46·2 (22)	70·6
	50	35·0 (22)	65·8
3	90	42·0 (30)	69·5
	50	33·7 (30)	64·2
4	90	42·2 (30)	72·4
	70	40·0 (34)	71·0
5	70	39·7 (30)	72·6
	50	32·3 (28)	68·3
6	90	42·6 (27)	72·5
	50	33·7 (30)	70·4

From these results it is clear that, in each of the six series, adults whose pupal period was spent in a higher humidity weighed more and had a higher water content than those from a lower humidity.

9. *The effect of temperature on the minimum relative humidity from which prepupae can absorb water.*

Previous experiments have shown that prepupae absorb water at 50 per cent. R.H. and above, but are unable to do so at 40 per cent. or below if the temperature is 24°C. The following experiment was carried out to define more closely the lower limit and to find whether this is affected by temperature.

Batches of about 30 freshly formed prepupae were put into squat pattern weighing bottles and kept at relative humidities of 35, 40, 45, 50 and 55 per cent., at each of the temperatures 12°, 24° and 35°C., and examined after three days to see whether they had dried up or appeared turgid.

At 12°C. the prepupae never pupated, but it was possible to see after three days whether they had dried up or not. At 35°C. all the prepupae which were going to pupate had done so after three days, but again it was possible to see whether they had dried up and shrivelled as larvae or pupated successfully.

The mechanical stimulus applied while collecting prepupae from the bedding caused a few to emerge from their newly formed cocoons, and some of these did not return. These naked prepupae have not been included in the results, but it is worth recording that they seemed to dry up more easily than those inside cocoons at the critical humidity.

The results (Table V) show that a relative humidity of 45 per cent. is the critical humidity at all temperatures used. Above this humidity absorption of water takes place normally, while below it desiccation occurs. It is very significant that it should be the same relative humidity at each temperature, since it shows that the process of absorption of water is not limited by any one saturation deficit. The results are somewhat at variance with those of Mellanby (1933) and will be discussed further below.

TABLE V.

The percentage, to the nearest 5 per cent., of batches of about 30 prepupae each, which avoided desiccation during the first three days of the cocoon period, at the temperature and humidities shown.

Temperature (°C.)	Relative humidity (percent.)				
	35	40	45	50	55
12	0	0	45	90	95
24	0	5	50	95	100
35	0	0	20	95	95

Discussion.

From the results of the foregoing experiments the water relations during the cocoon period can be summarised as follows. Within 24 hours of cocoon formation (and probably soon after defaecation) the prepupa begins to absorb water vapour from the air. It is capable of doing this whether silk and débris are present or not, though the presence of a cocoon allows the absorption of more water. Whether the decrease in amount of water absorbed when the cocoon is absent is due to increased activity on the part of the prepupa is uncertain. The process of absorption goes on until the prepupa moults and becomes a true pupa (which occurs about three and a half days after cocoon formation at 24°C.) and during this time prepupae increase their total weight by about 14 per cent., though the amount varies considerably and may be as low as 4 per cent. or as high as 29 per cent. During this period the dry weight of the insect falls by from 1 to 4 per cent. (mean about 2 per cent.) of the initial total weight, and since this is probably replaced by metabolic water, the net increase of 14 per cent. may be ascribed to water absorbed from the air. There is no indication that relative humidity or saturation deficit determines the amount of water absorbed, but there is a lower limit of relative humidity, which is about 45 per cent. at 12°, 24° and 35°C., below which prepupae are incapable of absorbing water and become desiccated. After true pupation, the insects begin to lose water and continue to do so throughout the pupal stage, and during this period the amount of water lost does appear to be related to humidity.

The change from feeding larva to prepupa is marked only by complete evacuation of the contents of the gut—no ecdysis occurs; and it would be interesting to know just when the faculty for absorbing water is developed. No experiments were made on feeding larvae in the present work, but Mellanby (1932a) has shown that whereas adult *Xenopsylla* can withstand exposure to a temperature of 38°C. for 24 hours at all humidities, the larvae can withstand only 22°C. in dry air and there is a regular rise in their thermal death point to 36°C. in 90 per cent. R.H. Bacot (1914), Buxton (1938) and others have found that low humidities adversely affect the larvae; so that the evidence suggests that the feeding larvae are water-losers. Even if this is so it is not necessary to assume a sudden change from the water-losing larva to the water-gaining prepupa, for the change may occur slowly during or towards the end of the last larval instar. The time when the change takes place and the mechanism of the change are subjects which deserve further study.

It is important to distinguish between a mere increase in percentage water content and an active intake of water vapour. The percentage of water in an insect's body may be increased by the retention of metabolic water. Mellanby (1934) for instance has shown that in 90 per cent. R.H., starved *Tineola* larvae increase their percentage of water by losing less water than the dry weight of food reserve oxidised. The proportion of dry matter to total weight fell in one batch from 40·7 per cent. to 37·0 per cent. in 14 days. Auber and Raymont (1944) working with *Ephestia* found that the water content of larvae reared at 30 per cent. R.H. was 57·3 per cent. and that pupae developed from them had a water content of 65·4 per cent. Fraenkel and Blewett (1944), though they obtained figures which differed somewhat from those of Auber and Raymont, showed that this rise in percentage water content occurred during the prepupal stage, and is the result of increased oxidation of dry materials at low humidities owing to an increase in the length of the prepupal period.

True absorption of water has been demonstrated in several insects. The eggs of *Gryllotalpa*, *Locusta*, *Dytiscus*, *Notostira* and other genera have been shown by various authors to absorb water, and Thompson and Bodine (1936) have shown that in *Melanoplus* the process is probably an active biological one and not due simply to osmosis, for the uptake varies with metabolic activity irrespective of the membranes present. Buxton (1932) suggests that the process is an active one in all these cases.

The eggs of all these insects are laid in contact with liquid water, but the uptake of water vapour itself has also been demonstrated; in *Chortophaga* by Ludwig (1937), in *Tenebrio* larvae by Mellanby (1932b), and in the ticks *Ixodes*, *Ornithodoros* and other genera, by Lees (1946); but in the examples quoted this is always accomplished in fairly high humidities. In *Chortophaga* the process only takes place above 82 per cent. R.H., and it is only at 96 per cent. R.H. that the total weight increases by as much as 15 per cent., and the uptake lasts no longer than 24 hours, after which weight is lost. In *Tenebrio* the uptake ceases below 88 per cent. In the tick *Ixodes* a point of equilibrium is reached at 92 per cent. R.H. and in *Ornithodoros* at a humidity a little lower than this.

As regards changes in composition during the prepupal and pupal periods in particular, Needham (1929) summarises the information available up to that date, from which it appears that the prepupae and pupae of different insects behave in very different ways. Some, such as the Sphingid moth *Deilephila* and the corpse-fly *Ophyra*, lose total weight, and the percentage of dry weight increases. Ludwig (1931) has also shown that the pupae of *Popillia* have a lower water content than their larvae. Others, such as the honey-bee and the silk-worm, although losing weight, increase the proportionate water content.

In no insect hitherto studied, so far as the writer's knowledge goes, has the prepupa been shown to absorb water from the air; and in those insects which have been shown in the larval or adult stage to absorb water vapour, the process does not occur in anything like the low humidities found in the present work.

Mellanby (1933) in an enquiry into the effect of temperature upon the lowest humidity required for pupation of *Xenopsylla cheopis* found that at temperatures from 18° to 29°C., although the duration of the prepupal period decreases with rise in temperature, the lowest relative humidity which allows satisfactory development of the larvae was constant at 60 per cent. At 35°C., however, he found that a R.H. of 75 per cent. was necessary. Expressing the humidity in terms of saturation deficit, he found that the product of this and the time necessary for pupation at each temperature was a constant, except at 35°C. where the product was lower than expected. Mellanby did not weigh his insects, but he argues that if prepupae lose water at a rate proportional to the saturation deficit of the air, they would be able to stand a high S.D. for a short time without losing so much water as to prevent pupation, and that this explains why the product : S.D. × Time is approximately constant.

The present results are nearly in agreement with Mellanby's as far as the observations at 18°–29°C. are concerned, for 45–50 per cent. is found to be the critical relative humidity, but do not agree with his failure to get pupation at 35°C. in 70 per cent. R.H. Mellanby remarks that he was unable to be sure that the larvae he used were fully fed, and that therefore the difference beteen 75 and 100 per cent. emergence is probably not significant. The prepupa (or fully fed larva) is distinguishable from the feeding larva by its empty alimentary canal, which gives it a white appearance, whereas feeding larvae are dark greyish brown, and the fact that Mellanby included some of these feeding larvae (? all feeding larvae) may account for the difference between his observations and the present results at 35°C.

The present experiments show that whereas the prepupae are indeed water-losers at 45 per cent. R.H. and below, they are water-gainers at all higher relative humidities. These results were obtained with *X. brasiliensis*, whereas Mellanby was working with *X. cheopis*, but it is submitted that two such closely related species are not likely to differ in their water relations to the extent that one is a water-loser in the prepupal period while the other is a water-gainer. Quantitatively, their water relations may differ slightly, and this may account for the somewhat lower R.H. found suitable for pupation in the present work ; but observations made on *X. cheopis* itself by the writer showed that the prepupa does increase in weight, though insufficient data are available to make the results worth recording in detail. It now appears that saturation deficiency does not come into the picture at all during the prepupal period, and that the only relation, provided the R.H. is 50 per cent. or above, is between time taken to pupate and temperature, which (as Mellanby shows) when graphed gives points lying close to a hyperbolic curve.

In other words, all the observations can be explained (except Mellanby's at 35°C.) without invoking saturation deficit and evaporation, and the observed facts do not provide evidence of the application of Dalton's law to the prepupae of fleas, as Johnson (1942), basing his remarks on Mellanby's results, suggests.

Once it has been demonstrated that the prepupae absorb water it is not surprising to find that the limiting factor for successful pupation is relative humidity and not saturation deficit, for the absorption of water vapour is known, in those cases where it occurs, to depend upon relative humidity (Mellanby 1932b).

Turning now to consider the behaviour of the insects after pupation has occurred, it is clear that they begin to lose water, for the total weight decreases (Table III) and the percentage water content also decreases (compare percentage water content at the end of the prepupal period in Table III with that of the freshly emerged adults in Table IV), and although a great deal of variation occurs in the amount lost, there is a very strong indication that more is lost in dry than in moist air. Evidence of this is provided by the weights of adults emerging from the different humidities, by their percentage water contents and indirectly by the small differences in longevity of such adults when starved and all kept at one humidity. The pupa is, however, capable of developing satisfactorily at 0 per cent. R.H. as Mellanby (1933) has shown, and it must, therefore, be capable of conserving its water efficiently, and this would account for the smallness of the differences in weight, water content and longevity found between adults emerging from various humidities.

The saturation deficits at 50, 70 and 90 per cent. R.H. and 24°C. are 10·8, 6·7 and 2·3 mm. Hg. respectively, and it is not possible with the small and variable differences found to say whether the loss of water during the pupal period is more closely related to relative humidity or to saturation deficit. On *a priori* grounds it is more likely to be related to saturation deficit.

As regards the mechanism by which water vapour is absorbed, not very much is known for certain. Various substances, such as paper and colloids, are known to adsorb water vapour from unsaturated air, the molecules then lose latent heat and enter the liquid state (Adam 1930). It is probable, therefore, that the prepupal cuticle is capable of adsorbing water in this way, and that the liquid water formed is then transferred into the body of the insect. On the mechanism of this process there is some difference of opinion. Mellanby suggests that it occurs through the tracheal endings, but Lees (1946) presents a considerable body of evidence showing that in ticks the absorption occurs through the cuticle by way of the pore canals, which are filled with cytoplasm in that animal. The cuticle of insects also contains pore canals, and in many species the canals are filled with cytoplasm. Wigglesworth (1933) has shown that they are so filled in *Tenebrio*, which is one of the insects which has been shown to absorb water vapour from the air. The canals do not penetrate the thin epicuticle, but Lees suggests that their tufted external ends, exposing as they do a very large surface of cytoplasm, may well actively absorb water from the epicuticle, and the other (internal) ends secrete water into the haemocoele. Wigglesworth has also pointed out that where the pore canals are filled with cytoplasmic processes from the epidermal cells immediately below the cuticle, this would enable the cells to exert their effect directly upon the exo- and epi-cuticles, by the passage of substances synthesised in the cell body.

In fleas the present experiments have shown that there is a sudden change in behaviour as regards water absorption at the time of pupation. There may also be a sudden change between the behaviour of feeding larvae and prepupae in this respect, though the possibility must not be overlooked that feeding larvae also absorb water from the air in the same way as prepupae, but get rid of the excess by excretion through the alimentary canal. In the change from prepupa to pupa there is a change in cuticle, whilst from feeding larva to prepupa there is not. The explanation in the first case may therefore be that the prepupal and pupal cuticles are constructed differently, the one to allow water to pass easily in an inward direction, the other to obstruct the flow of water in either direction ; and the explanation of the second case may be that the necessary changes in the structure and properties of the cuticle are brought about by the action of the epidermal cells through their cytoplasmic processes in the pore canals.

In this connection the work of Hurst (1941) is of interest. He and other workers have shown that double (asymmetrical) membranes are frequently much more permeable to a given substance in one direction than in the other, that the permeability is by no means equal to the mean of the permeabilities of the two membranes considered singly, and that the permeability may be altered to a very great extent by the relative distribution and composition of soluble and insoluble components of the membranes.

The structure and physical properties of the cuticle of flea prepupae and pupae have not been studied, but there is little doubt that such an enquiry would yield interesting information.

Summary.

1. The original purpose of the present work was to confirm a previous finding that, in unfed adult *Xenopsylla*, longevity varies according to the humidity during the cocoon period, and to find whether this is due to differential loss of water during

the prepupal period. The previous finding has been confirmed, but the cause is shown to lie in loss of water during the pupal and not the prepupal period.

2. Prepupae are shown to gain in weight by the absorption of water vapour from the air if kept at 24°C. and any relative humidity from 50–90 per cent. If kept below 45 per cent. R.H. they rapidly dry and fail to pupate.

3. During this period (about three and a half days from cocoon spinning to pupation at 24°C.) the dry weight decreases by about 2 per cent. of the original total weight, and the total weight increases by from 4–29 per cent., with a mean of 14 per cent.

4. The amount of water absorbed is independent of relative humidity within the range 50–90 per cent., and is approximately equal to the increase in total weight.

5. The critical relative humidity, below which the insects die and above which they absorb water, is shown to be substantially the same (from 45 to 50 per cent. R.H.) at 12°, 24° and 35°C.

6. After pupation the insects begin to lose water and continue to do so throughout the period. The rate of loss increases with decreasing humidity, though the effect of humidity is not very great.

7. Adults emerging from pupae kept at higher humidities weigh more, have higher percentage water contents, and live longer without food, than those emerging from pupae kept at lower humidities.

8. The results are discussed in the light of previous work, and suggestions as to the mechanism of the process of absorption of water are made.

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C

A NEW SPECIES OF ENCYRTIDAE PARASITIC IN *COCCUS HESPERIDUM*, L.

E.H.N. T.O

By Harold Compere.

Associate in the Experiment Station, University of California, Citrus Experiment Station, Riverside, California.

(Plate II.)

The species described as new is placed in the genus *Adelencyrtus*, Ashmead, provisionally. The description is published at this time to provide a name, so that biological information concerning the species can be made a matter of record. The new species is most closely allied to *Adelencyrtus*. It differs from typical species of this genus with regard to the mandibles. In typical *Adelencyrtus* the mandibles are quadridentate, and the ventral margin is provided with a stout, cone-shaped seta. In the new species only the ventral tooth is distinct ; the dorsal margin of the cutting edge appears as a wide truncation, which can scarcely be described as subdivided, and the ventral margin of the mandible lacks the stout conical seta. The most useful character for the identification of this species, and the one which reveals its relationship to *Adelencyrtus* most surely, is the distinctive male antennae. In this sex the antennal club is enormously cylindrically elongated, and the funicle is composed of two vestigial segments that are contained in a recess at the base of the club. Antennae of a similar kind have been described in the males of *Habrolepis*, Förster, and *Anabrolepis*, Timberlake, two genera which are closely allied to *Adelencyrtus*. The males of *Adelencyrtus* have not been described.

In the collection of the University of California Citrus Experiment Station are specimens of an undescribed species of *Adelencyrtus*, with quadridentate mandibles, collected by E. W. Rust in Natal, South Africa. This species is represented by both sexes. In the males the antennal club is cylindrically elongated, and the funicle composed of two vestigial segments.

In the females the following major characters indicate the relationship of the new species to *Adelencyrtus* : thorax relatively shallow and flattened above ; face inclined ventro caudad ; head convex above, the dorsal and anterior planes meeting in a wide curve. The habitus resembles that of a parasite inhabiting a Diaspine scale more than that of a parasite inhabiting a Lecaniine scale.

With regard to host relationships, the undescribed species from Natal, previously mentioned, which almost certainly belongs in *Adelencyrtus*, is recorded by Rust as having been reared from *Inglisia* sp. and *Ceroplastes* sp. It is unusual to find species of the same genus parasitising both armoured and unarmoured scales. The greater number of species of *Adelencyrtus* parasitise the armoured Diaspinae.

Adelencyrtus simmondsi, sp. n. (Pl. II ; fig. 1[*]).

♀. Greater part of head, and the axillae and scutellum, bright bluish green ; in some specimens mesoscutum concolorous with scutellum and head ; in others mesoscutum more strongly bluish or purplish. Basal tergum of abdomen bright metallic, the sides bluish green. Metanotum and propodeum black. Sides and under parts of thorax dark brown. Abdomen except as noted, blackish and faintly metallic. Face between antennae grading to metallic blackish. Antennae pale straw coloured with the club more brownish. Legs largely white to pale straw coloured with the hind tibiae black except the ends. Mouth parts white except the mandibles. Front wings with a wide, dark band across the middle; the banded effect produced by crowded, coarse, dark hairs in addition to some infuscation of the integument; basal and apical areas of the wing clothed with fine, whitish hairs.

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[*] The figures of the adults were prepared by Hubert W. Simmonds.

Mesoscutum with numerous small, squamous white hairs. Eyes almost imperceptibly hairy. Frontovertex, face, and cheeks with fine refractive hairs. Sides of propodeum with tufts of white hairs.

Frontovertex, temples, cheeks, axillae and scutellum punctate reticulate; mesoscutum less distinctly punctate reticulate, in slight contrast to the punctate areas enumerated. On the frontovertex the fine hairs arise from punctations that are slightly larger than the reticulate punctations.

Head rounded above, frontovertex and face meeting in a wide curve. Frontovertex narrow, fully twice as long as wide. Ocelli in an acute triangle, the anterior ocellus about once its own diameter from the orbits, and the posterior ocelli about two diameters from the occipital line. Scrobes triangular, meeting above. Eyes short, widely divergent below. Mandibles, as shown in fig. 1; a small ventral tooth and a wide dorsal truncation; the basal margin of the truncation is very slightly produced, scarcely forming a tooth. Maxillary palpus four segmented; labial palpus three segmented. Antennae relatively short; pedicel twice as long as wide, longer than the two succeeding segments; first five segments of the funicle equal or nearly in length, and very slightly increasing in width; the first about as long as wide; the fifth appreciably wider than long; sixth segment of the funicle disproportionately enlarged relative to the preceding segments, almost twice as wide as the first but scarcely much longer; club ovate, about twice as long as wide, and twice as wide as the preceding segment.

The marginal, postmarginal and stigmal veins are partly concealed by covering coarse dark hairs, and infuscations. Postmarginal and stigmal veins of equal length; marginal vein at least as long or longer than the stigmal or postmarginal veins.

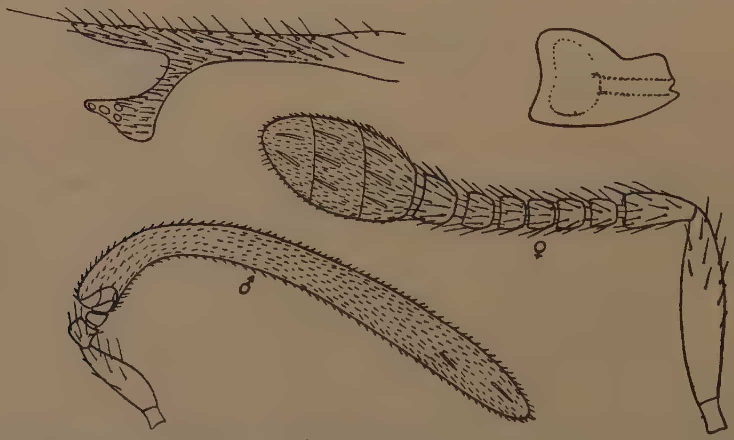

Fig. 1.—*Adelencyrtus simmondsi*, sp.n. Marginal, post marginal and stigmal veins; mandible; male and female antennae.

♂. Head largely bluish green; punctate reticulate. Mesoscutum, axillae and scutellum, dark metallic, with variable bright reflections depending on the light. Antennae white to pale straw coloured with the pedicel suffused with brown. Legs largely white; hind femora faintly marked with dilute brown or blackish near the middle; hind femora blackish except the ends; hind tarsi brown. Mesoscutum with a covering of white hairs. Front wings without a cross band. Antennae as shown in fig. 1.

Described from a mixed lot of specimens, 14♀♀ and 10♂♂ received from the late Professor James Davidson, Waite Agricultural Institute, South Australia, labelled as follows : 1 ♀ " ex *Coccus hesperidum*, Simmonds Coll., sp.n.; 2 ♂♂ ex *C. hesperidum*, Simmonds Coll. Par. O.; 7 ♂♂ Series 13° ex *C. hesperidum*, Netherby, South Australia, 28/x/44 ; 7 ♀♀ and 1 ♂, ex *C. hesperidum*, Series 11 D, H.W.S. The specimen enumerated first is the holotype ; a specimen of the lot labelled " Series iii 13° " is selected as the allotype ; the remainder of the specimens are designated as paratypes.

Types to be deposited in the British Museum (Natural History).

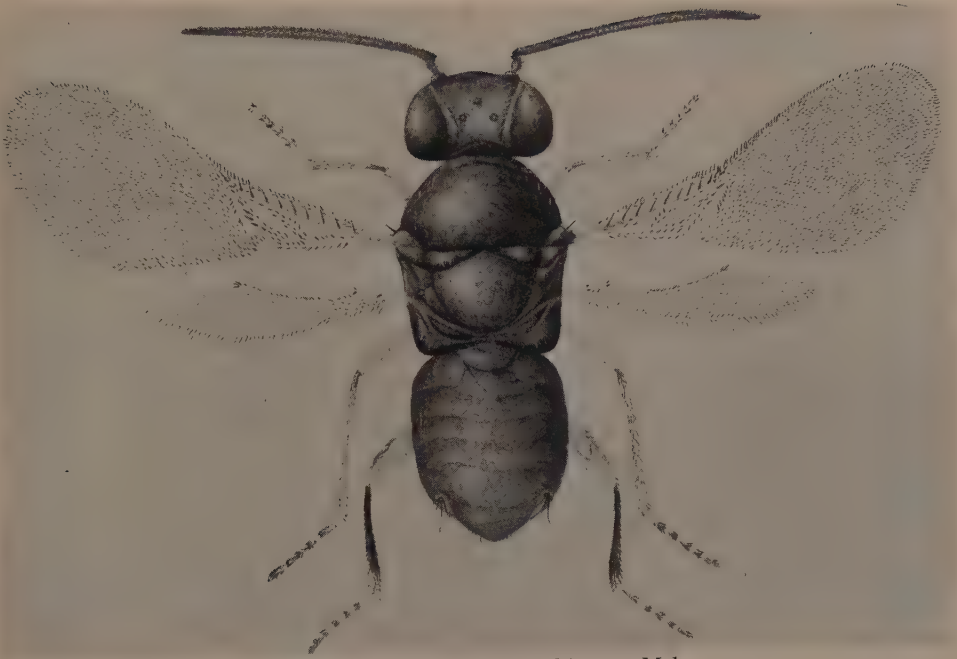

FIG. 1. *Adelencyrtus simmondsi*, n.sp. Male.

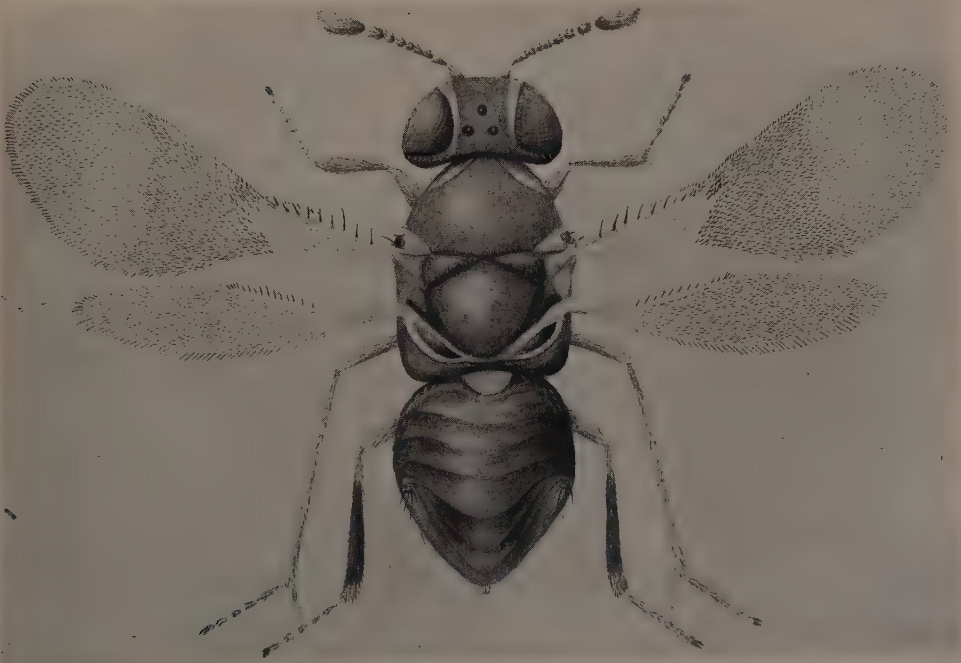

FIG. 2. *Adelencyrtus simmondsi*, n.sp. Female.

THE CLIMATOLOGY OF BLOWFLY MYIASIS.

I.—Weather and Oviposition.

By John MacLeod.

Cooper Technical Bureau, Berkhamsted.

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General Introduction.

The study of blowfly-climate relationships is much complicated by the multiplicity of factors involved. The observed relations of the three main components, climate, blowfly and sheep, may be due to almost any combination of the factors into which these components divide. The factors may be listed thus :—

Climate.

(*a*) Broad differences in facies.
(*b*) Periodic variations—seasons.
(*c*) Irregular variations—weather.

Blowfly.

(*d*) Population.
(*e*) Activity.
(*f*) Response to sheep odours.
(*g*) Viability of eggs deposited in fleece.

Sheep.

(*h*) Fleece type.
(*k*) Moisture balance in fleece.
(*l*) Development of attractant odours.

Response to attractant odour, *i.e.* oviposition, is not necessarily synonymous with fly activity, for two reasons: it is a specialised activity by a special section—the gravid females, and also it is possible that weather conditions may influence the effective range of odour and/or the sensitivity of the receptor organs concerned.

The principal interrelations of the above groups of factors might be expected to operate somewhat after this fashion :—

(*a*) affects (*d*) but this is also dependent on (*b*).
(*a*) ,, (*h*).
(*b*) ,, (*e*) ,, ,, ,, ,, ,, ,, (*d*) and (*c*).
(*b*) ,, (*k*) ,, ,, ,, ,, ,, ,, (*h*) ,, (*c*).
(*c*) ,, (*l*) ,, ,, ,, ,, ,, ,, (*k*) ,, (*h*).
(*c*) may possibly affect (*f*), but this principally dependent on (*e*).

A variation in oviposition may be due to (*b*), or (*c*), operating through (*e*), to (*f*) independently of (*e*) or to (*l*).

The general investigation of blowfly myiasis and climate thus falls into three main divisions—(1) a purely autecological study of the blowfly, in particular its population structure and density in relation to weather, (2) the dynamics of the fleece, in particular the intereffects of fleece type, weather, and decomposition processes, and (3) the relation of climate to blow and strike. Only this last aspect is dealt with in the present series of studies.

Analysis of the Problem.

Field evidence of incidence of strike in relation to weather, although it cannot be interpreted directly, will nevertheless often be the only evidence available, because of the impracticability of experimental analysis. It is necessary, therefore, to have clearly in mind the intereffects involved. Weather and myiasis show four relationships, which may not be identical—the effect of weather (1) on fly availability (activity), (2) on the development of attractant odours in the fleece, (3) on the response of flies to these odours, and (4) on the fate of deposited eggs. It may not always be possible to separate these intereffects by analysis (*e.g.* activity and response to odours), but it should be recognized that they exist.

Generally speaking, the different types of evidence may be interpreted as follows:—

Where sheep are "attractivated" by deliberate addition of a source of attractant odour, or with baited trap results, variations in "blow" (*i.e.* in oviposition) may be directly interpreted as fly response, or fly response modified by the activity variable (1).

Where the flock is subject to natural conditions, *i.e.* the sheep not attractivated, variations in observed blow may be a direct expression of fly response, or of fly response modified by activity and/or development of attractant odours (2). Differences between treated and control flocks will give evidence on this (2) variable.

The distribution of naturally occurring strikes ("strike" means the successful development of larvae from deposited eggs, resulting in a skin lesion), where the date of oviposition is estimated from the state of the strike lesion, may be regarded as a measure of the weather effect on viability of deposited eggs (4), conditioned by the degree of oviposition (3), which in turn may be an intereffect of (3) and (2)—development of odour, (3) being further qualified by variation of (1)—fly activity. Difference from distribution of natural blow will give evidence on viability (4).

Separate records of naturally occurring strike and observed blow, and experimental records of blow in response to attractants, adequately controlled, should thus provide data for a fairly complete analysis of the relation of weather to myiasis. Adequate records of natural blow have not been obtained, but strike records over a number of years at the Research Farm, similar records of experimentally induced blow, and records during one blowfly season of the relationship of oviposition to actual, as opposed to daily average or maximum, climatic element values, form the basis of the present study. In this first paper the exact data on weather elements and oviposition are analysed as a preliminary to the interpretation of results based on daily meteorological records.

Methods.

The experiment was carried out in the summer and autumn of 1942, at the Cooper Field Research Station, Herts. The same three Hampshire cross lambs were used throughout the test. Pledgets of cottonwool, about 1 in. × $\frac{1}{2}$ in., made to a standard pattern were soaked in fresh 4 per cent. ammonium carbonate solution and sewn to the wool in the region of the loins. An hour later the egg clusters were noted and removed, and a fresh pledget attached. Meteorological readings were made at the beginning and end of each one hour period. From 4 to 7 successive tests were made on each day, giving a total of 134 one-hour exposures. The lambs remained in the same paddock throughout the experiment. A sample from each egg cluster was cultured as a check on the species.

The meteorological station was about 100 yards away. Readings were made of dry and wet bulb temperatures in a Stevenson screen, and of sun temperatures from a black bulb *in vacuo* thermometer in the open. The difference has been taken as a measure of intensity of solar and sky radiation. Evaporation was measured by a Livingstone Standardized White Atmometer Sphere, mounted about 15 ins. above the grass surface.

Unless otherwise stated, the temperature and air-humidity readings used are the means of the initial and final readings for the hour ; the atmometer is, of course, self-integrating, and the figure used is the evaporation in ccs. for the test hour. The correction coefficient was ·85, but its application in this series of readings would be superfluous.

As a check on the possibility of a time lag with the enclosed thermometers, simultaneous readings of air-temperature and humidity were also made from a whirling hygrometer. No consistent significant difference was obtained and only the screen readings have been used.

Homogeneity checks.

It is known that there are considerable differences in the susceptibility of sheep to blow. These differences may persist even when the general level of susceptibility is raised by attractivation, and this source of variability was, therefore, excluded by the use of the same sheep throughout. It has been found, however, that susceptibility varies, not only from sheep to sheep, but in the same sheep at different times, even when attractants are used. This is shown by the records for two series of tests on a group of five sheep, both during 1936 (Table I).

TABLE I.

Sheep	Series 1							Series 2						
670	+	−	+	+	+	+	5/6	+	+	−	+	+	−	4/6
690	−	−	+	+	+	−	3/6	−	−	−	−	−	+	1/6
669	+	+	+	+	−	−	4/6	+	−	−	−	−	+	2/6
676	−	−	−	−	+	−	1/6	−	+	+	+	−	+	4/6
653	+	−	+	+	+	−	4/6	+	−	−	−	−	−	1/6

The result illustrates both the variation between sheep under the same conditions, and the variation in relative susceptibility of given sheep at different times. Thus, sheep 676 and 653 have reversed their relative susceptibilities between the two series.

This variability is uncontrollable, but, as it happened, the three sheep remained a fairly homogeneous group, and their relative susceptibility order remained unchanged. The total figures were :—

Sheep M 42, 24	positives	51	egg clusters.
,, M 43, 26	,,	65	,, ,,
,, M 44, 33	,,	74	,, ,,

Species check.

The species of fly striking sheep at the Research Station was known to be almost exclusively *Lucilia sericata*, Mg., other species rarely occurring in strikes. Whether this was a reflection of the relative blow incidence, or whether it was due to a differential mortality of eggs, was not known, but in this experiment the first explanation would appear to be true. Of the 190 egg clusters obtained, 174 were tested for identity ; of these, 5, all in July and early August, were sterile. The remainder, with one exception were *L. sericata*. The exception, one cluster of *L. caesar*, L., eggs, occurred on the 11th August, along with 16 *L. sericata* clusters.

Preliminary Experiment.

Before detailing the results of the 1942 experiment, it may be of interest to give the results of a small scale test carried out in 1936, as these formed the basis of expectation for blowfly work in the intervening years ; they also gave the first indication that, contrary to prevalent belief, humidity might not be positively related to blowfly activity, *i.e.* that muggy days might not be more favourable for fly activity than bright days. Three sheep were attractivated and exposed for periods of 1 to 3 hours, on nine occasions during the latter half of August. The results were expressed in terms of oviposition per hour, ranked and tested for correlation. Spearman's ranked correlation value for meteorological factors is :—

Air temperature	·64	Relative humidity	—·51
Sun temperature	·5	Saturation deficiency	·63
Diff. (=radiation)	·08	Evaporativity	·6

The absence of correlation of "activity" with solar radiation, and the closer correlation with air-temperature than with temperature in the sun, are interesting. Activity and moisture content of the air appeared to be inversely related ; the relation was better for humidity expressed as saturation deficiency than for percentage saturation.

The 1942 Experiment.

General observations.

The tests were spread over the period 28th July to 16th October (see fig. 2). No oviposition occurred after the 9th October. It is improbable that this is a phenological cessation of fly activity, for blow has been obtained experimentally, or observed in nature later in the season, in 1936, 1937, 1938 and 1939. It is shown in fig. 2 A that from 9th October onwards the temperature remained consistently below the lowest point at which blow was obtained in the experiment. Nevertheless, since this is near the end of the blowfly season, the possibility of a seasonal effect could not be dismissed, and the results have, therefore, been examined both with and without those obtained from 9th October onwards.

The distribution of tests and results over time of day (G.M.T.) is shown in Table II.

TABLE II.

Time ...	8–9	9–10	10–11	11–12	12–1	1–2	2–3	3–4
Number of tests	5	14	23	19	21	22	20	10
Number of negatives	5	13	17	15	13	13	9	6
Number of egg clusters	0	1	18	24	63	38	39	7
Ratio of clusters to tests :—								
All results	43 : 61				147 : 73			
Excluding late tests	43 : 51				147 : 53			

There was, over the period of test, a greater degree of blow in the afternoon than in the forenoon.

The frequency distribution of oviposition was as follows :—

Clusters	0	1	2	3	4	5	6	7	8	9	14	16	17	24	Total
Frequency ...	91	14	5	8	4	1	3	1	2	1	1	1	1	1	134

The curve is excessively biassed so that the usual parameters, the mean and variance, would have little descriptive value. The log values, with unity added to each result, have, therefore, been used, to render the skewness less extreme. The curve is still, however, abnormal and the geometric mean is less than the unit of measurement.

As a preliminary to the detailed examination of each separate factor, the mean of readings of each for tests giving high counts (6 clusters or over) was compared with the general mean. Since the two samples are not independent, the standard error was taken as the square root of the variance of the main sample divided by eleven, the size of the subsample (Table III).

TABLE III.

	Temp.	Sun Temp.	Rel. humidity	Evaporation
Subsample mean	65·136	98·273	63·455	1·773
Population „	60·165	88·62	70·037	1·474
Difference	4·97	9·65	6·58	0·3
Standard error	1·42	4·69	3·21	0·27

The first three factors have a significantly higher mean value for those tests which gave good results, indicating a positive correlation ; the evaporativity index is also higher but the difference is not significant.

Air temperature.

The distribution of temperature is reasonably normal :

Temperature	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	68	69	70	71
Frequency all results ...	2	2	9	3	15	6	5	7	7	16	14	11	9	4	5	6	3	4	3	1	2
Frequency—excluding late tests ...	0	1	1	0	7	2	3	5	7	16	14	11	9	4	5	6	3	4	3	1	2

The dependence of degree of oviposition on temperature is obvious, as shown in fig. 1. The correlation coefficient for all results is ·586, with a standard error of under ·1. The regression of oviposition on temperature has a coefficient of ·042, so that the log of one plus the number of egg clusters increases by ·3 for a rise of about 7·5°F. That is, the number of egg clusters (plus one) would be twice as high.

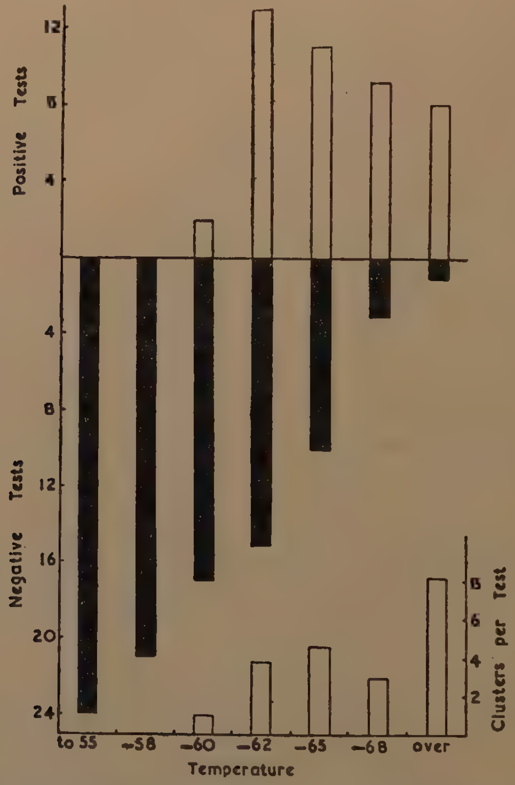

Fig. 1.—The number of tests at each temperature interval, divided into: positive tests (above the line) and negative tests (below). The average number of clusters for the positive tests is shown in the bottom part of the diagram.

Analysis of the variance for all results shows no significant departure of the regression from linearity.

The regression line cuts the x axis at about 56°F. The lowest temperature at which oviposition occurred was 58°—the mean of 56·5° and 59·5°, initial and final temperatures. There were 42 tests with mean temperatures below 58°. Blow can occur at temperatures below 56°, but usually only in a very low proportion of exposures, (on the basis of this experiment the expectation would be about 2 per cent.), so that for general purposes the lower temperature limit has been taken as about 58°F., or 15°C. The late season group of tests, with one exception, gave readings below this (fig. 2 A), so that the simplest explanation of the absence of positive results in the group is that of temperature control.

The effect of temperature history of the previous one and two hours on the result of the test hour was examined by plotting the oviposition results on charts with ordinates max. experimental temperature and total temperature increment for the two and three hours respectively. The absence of instances of test-hours near the

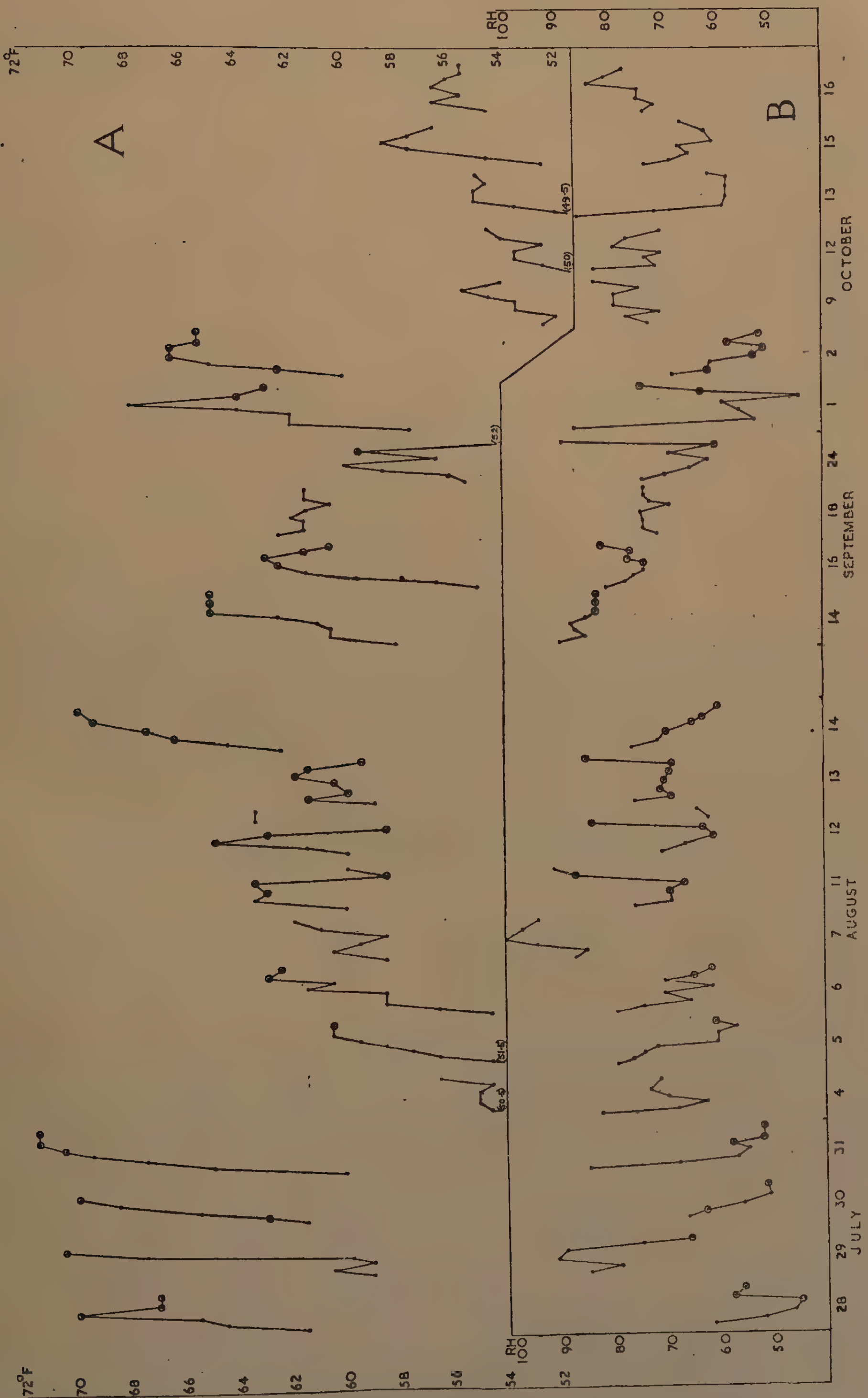

Fig. 2.—Showing the daily sequence of tests in relation to (A) air temperature, and (B) relative humidity. Test hours during which oviposition occurred are marked with large dots for the terminal reading.

lower critical limit with a history of a warm preceding hour is unfortunate, as it would have been interesting to find whether the lower critical temperature is affected by such history. For many of the negative tests there is no record of the previous two hours. The nine available negative tests at warm temperatures, 64° and over, all had histories of a distinctly cooler preceding hour :—

Test hour max.	64	65	65·5	67·5	68	68·5	69·5
Preceding hour mean ...	62	60	62·5	61·5	61·5	64·5	65·5
			61·5	55·5			

Otherwise, the chart failed to reveal any relation, because of the close correlation of the two variates, giving in this experiment remarkably little dispersion. The comparison with the two previous hours gave rather more scatter of results, but showed less association.

Solar Radiation.

The frequency distribution of temperature in the sun, although not markedly skewed, gave a much lower and broader curve than that of temperature—the standard deviation is over one-quarter of the total range.

	60	65	70	75	80	85	90	95	100	105	110	115	120
Frequency ...	3	7	11	18	17	12	12	14	8	12	10	6	4
Positives ...	0	0	0	3	5	5	4	5	3	7	4	4	3
Clusters per test	—	—	—	0·33	1·9	1·25	1·4	0·6	1·0	4·5	0·9	4·5	3·5

The correlation here is much less obvious. Calculated in 5° intervals of T_s (sun temperature) r is ·37 and the regression, b, =·008 for each degree. Solar radiation, though tending to be high with high air temperature, is by no means exactly correlated. There is a considerable scatter of individual results (r=·44) : It has, therefore, been examined independently of air temperature. The distribution is reasonably normal :—

0–5	6–10	11–15	16–20	21–25	26–30	31–35	36–40	41–45	46–50	51–55
1	10	12	20	17	12	16	14	11	11	6

r=·218+0·087 ; b, for 1 degree,=·0054, *i.e.* the mean value for n+1 clusters would double for a rise of 55° in $T_s - T$.

Humidity.

The relation of relative humidity to oviposition over the test period is illustrated in fig. 2 B. The mean relative humidity is 70·04. Ignoring the initial readings for each series of tests, there are 63 readings above the mean, 20·5 per cent. of them being positive ; 71 below have 42 per cent. positives. Omitting the late tests the percentage positives above and below the 70 R.H. line are 29·5 and 50. For the effective season tests the results, compared with those to be expected from a random distribution, are :—

	Under 60 R.H.	60–69	70–79	80+
Readings ...	22	36	26	21
Positives ...	11	18	7	7
Expected ...	9·1	14·45	10·75	8·7

The total (unadjusted) χ^2 for the comparison is, however, only 4·9, which, with 3 degrees of freedom, is not significant.

The R.H. distribution is the most normal of all the independent variables. With 5 per cent. intervals, it is :—

R.H. ...	45	50	55	60	65	70	75	80	85	90	95	100
Frequency	1	6	12	13	20	24	30	12	10	3	2	1

The correlation with log oviposition is significant (r=—·249±·087). The regression of oviposition on R.H. is —·008. The group means for each 5 units of humidity suggest that the regression line rises more steeply at the lower humidities, but analysis shows that the variance of the group means from the regression line is considerably less than the error variance.

The humidity results were also examined as millibars deficit from saturation vapour pressure. The distribution in millibar units, is :—

0·5	1·5	2·5	3·5	4·5	5·5	6·5	7·5	8·5	9·5	10·5	11·5	12·5
3	1	10	26	17	28	17	8	6	4	8	3	3

The correlation is closer than for R.H. (r=·406), but still considerably scattered. b=·051 for each millibar, *i.e.* about 12·5 per cent. It should be remembered, however, that for a given percentage saturation, saturation deficit is a function of temperature, which has a fairly high correlation with oviposition. The correlation between saturation deficit and air temperature in this experiment is, in fact, very high (r=·71). The effect of this on the oviposition and saturation deficit correlation is examined later.

Drying Power of the Air.

The rate of evaporation from a white atmometer sphere is an integration of the effect of air temperature, humidity, and wind, but not of radiation. The distribution is :—

ccs.	0	0·5	1·0	1·5	2·0	2·5	3·0	3·5	4·0	4·5	5·0
	8	10	48	22	30	4	8	0	2	1	1

r=·27±·087.

b=·1, and since the correction factor for the instrument used is ·85 this represents an increase of the value N+1 egg clusters of about 31 per cent. for 1 cc. increase in evaporation per hour by the standard Livingstone atmometer.

Wind.

No direct measure of wind velocity was made, and this omission is to be regretted since wind is obviously unfavourable, both for mechanical reasons and, if continuous, by reason of the consequent inability of the attracted fly to locate the source of attraction. It is much to be desired, therefore, that the limiting wind strength for fly activity and for oviposition—the values may not be identical—should be determined. The indirect assessment of the significance of wind in this experiment is discussed later.

Precipitation.

Rain showers occurred fairly frequently during the test hours, and on some of these occasions positive results were recorded at the end of the hour. With continuous rain, however, no oviposition was obtained, and there is not sufficient evidence on which to examine the effect of recent rain on oviposition, with the other factors eliminated.

Intereffects due to Independent Variations of Meteorological Factors.

Solar radiation and temperature.

It might, *prima facie*, be anticipated that the two effects would be additive. Failure of sun temperature to give a higher correlation than air temperature does not negative the possibility, because temperature in the sun does not have a fixed ratio of air-temperature and insolation. Examination of the data, however, revealed an unexpected antagonism of effect—high insolation, for a given temperature, tends to decrease the oviposition rate.

The corresponding solar radiation was plotted against the higher of the two air temperature readings for each hour, and the result entered at the plot point. This was considered preferable to the use of the mean values, since there is no evidence that the pairs of mean variates occurred simultaneously. The temperature range was divided into a series of fairly close intervals, and a natural division near the middle of the scatter of results for each interval was taken as a working median insolation. The proportion of clusters above and below the median insolation for each temperature interval was then noted. The mean number of clusters above and below the medians, and also the geometric means, which reduce the effect of single large values, were calculated (Table IV).

TABLE IV.

Temp.	No. of tests		No. of clusters		A.M.		Summed log. $N+1$		Mean log.		G.M.	
	Above	Below	Above	Below			Above	Below				
60–61	11	14	4	4	0·36	0·29	1·08	1·08	0·098	0·077	0·25	0·19
62–63	13	9	18	40	1·38	4·44	3·86	4·44	0·297	0·493	0·98	2·11
64–65	7	7	17	13	2·43	1·86	1·78	2·6	0·254	0·371	0·79	1·35
66–68	6	5	6	12	1·0	2·4	1·5	1·9	0·25	0·38	0·78	1·4
69–71	5	5	24	48	4·8	9·6	3·7	4·01	0·74	0·802	4·5	5·34

Because of the abnormal distribution of the oviposition frequencies the comparisons of these pairs of means by using the standard error of their difference was not considered legitimate. The first and second, and the third and fourth rows have been combined, and the overall association tested for significance (Table V).

TABLE V.

Temp.	S.R.	No. of tests	Frequency of clusters		Contributions to χ^2
			Expected	Observed	
Below 64	High	24	33·7	22	4·06
	Low	23	32·3	44	4·24
64–68	High	13	24·9	23	0·14
	Low	12	23·1	25	0·16
69–71	High	5	36·0	24	4·0
	Low	5	36·0	48	4·0

The total χ^2 value is 16·6, and since each temperature range has one degree of freedom, the probability of random distribution is less than 1 in 100 chances.

It was expected that near the lower critical limit of temperature insolation might increase activity, but the examination showed a complete absence of any tendency for the lower limit to be depressed by insolation.

Humidity and temperature.

Examination of the ratios of ovipositions above and below median humidities for given temperatures showed a slight tendency for oviposition to be inversely related to the degree of saturation. The association is insignificant, P′ being just under ·5.

TABLE VI.

Temp. Intervals	High R.H.			Low R.H.			Total χ^2
	Over 5 clusters	1–5	0	Over 5 clusters	1–5	0	
60–61	2	2	11	1	7	7	
62–63	0	3	7	2	4	4	
64–65	0	4	0	1	1	3	
66–68	1	1	1	1	2	1	
69–71	1	2	0	1	2	0	
Total	4	12	19	6	16	15	
Expected	4·86	13·6	16·5	5·14	14·4	17·5	
χ^2	0·152	0·188	0·378	0·144	0·178	0·357	1·397
	High evaporation			Low evaporation			
60–61	0	8	8	3	2	9	
62–63	2	4	2	0	0	7	
64–65	0	1	3	1	2	0	
66–68	1	2	1	2	1	2	
69–71	2	1	0	0	3	0	
Total	5	16	14	6	8	18	
Expected	5·75	12·55	16·7	5·25	11·45	15·3	
χ^2	0·098	0·949	0·437	0·107	1·039	0·476	3·106

The fact that the tendency was in this rather than the opposite direction was unexpected, in view of the insolation temperature intereffect, so the evaporation rate, as an independent estimate of air moisture, was examined in the same way. Because of the limited number of groups in the evaporation range, it was not always possible to divide the scatter for each temperature interval into approximately equal halves, and some results, on the median evaporation, have had to be omitted.

As with R.H., evaporation for a given temperature tended to be positively related to oviposition, suggesting an inverse air-humidity relation. The results (Table VI), show that for high oviposition values the relationship is reversed. If, however, the totals are treated as a $2 \times n$ contingency table, the total contribution of this aberration to the χ^2 for the table is only ·205 out of a total 3·106. The overall association is again insignificant, but with a lower probability of randomness than for R.H.—about ·3. The product of these two independent estimates of the humidity effect is also insignificant.

Treatment of the sun temperature data also showed no intereffect of humidity and sun temperature.

Evaporation and insolation.

The above method was also applied to examine the effect of air-drying power on the relation of oviposition to solar radiation. The results, for tests above the lower critical temperature limit, are given in Table VII. The total χ^2 is 9·38, which with 2 degrees of freedom has a probability level of under ·01.

TABLE VII.

T_s–T range	High evaporation			Low evaporation		
	Over 5 clusters per test	1–5	0	Over 5	1–5	0
Under 20	0	4	1	1	0	7
20–32	2	8	3	2	3	7
33–40	1	2	5	2	3	4
41–44	1	3	0	0	1	3
45 +	2	3	2	0	3	5
Total	6	20	11	5	10	26

There is thus a definite association of evaporation with the oviposition response to insolation, high evaporation increasing the rate of oviposition for a given intensity of insolation.

For a given insolation range evaporation values below the median may be due to the drying power of the air actually being low, or alternatively to insolation itself being responsible for much of the drying power. This component of the air drying power would not be registered by the atmometer. The method, therefore, only tests the influence of the drying power due to the remaining components, temperature, humidity and wind, on the oviposition relationship. Since humidity and insolation are not independent, the analysis resolves itself into the influence of the drying power of the air due to temperature and wind on the insolation effect. Much of the above significant association is, therefore, probably ascribable to temperature.

Intereffects due to related Variations of Meteorological Factors.

A factor, A, can influence the response of oviposition to another factor, B, in two ways. Independent variations of A can alter the result of B variations. These are the intereffects discussed in the foregoing section. Thus, insolation influenced the relation of oviposition to air-temperature, and evaporation variations varied the effect of insolation on oviposition. In addition, factor A may have an effect on the result through being itself related to factor B. A variation in B will thus mean coincident variation in A, and if A is itself influencing oviposition, the oviposition response will necessarily reflect the influence not only of B's variation, but also of A's simultaneous variation. The strength of such indirect influence of A can be assessed by comparing the crude correlation of B with its partial correlation where the effect of A's coincident variations are eliminated.

The two types of intereffect should not be confused; the second is due to simultaneous and related variations of two "independent" or x factors, and is predictable, whereas the first is due to independent variations of one x factor with regard to the other, and consequently its incidence will be erratic.

It is also possible to eliminate, by mathematical treatment, the influence of two or more related factors from the result to be credited to the factor under consideration. The method has been applied in some detail to the interrelations of oviposition and the three factors air temperature, insolation and moisture content of the air.

Interdependence of meteorological factors.

The correlations of the different combinations of meteorological factors are given in Table VIII.

TABLE VIII.

	Total correlations				Partial correlations		
	S.R.	R.H.	S.D.	E.	Corr.	Eliminated	Partial r.
T	0·44	0·44	0·71	0·45	T.SD	S.R.	0·63
T_s			0·73	0·61	T.SR	S.D.	−0·05
SR			0·66	0·53	SR.SD	T	0·54
SD				0·65	T_sSD	E	0·56
					T_sE	SD	0·27
					E.SD	T_s	0·37

All the meteorological factors discussed are related more or less closely. The reason for some of the relationships is obvious, *e.g.* temperature and degree of saturation, since temperature determines the vapour capacity of the air. Others are indirect, *e.g.* temperature and insolation. Water is the main absorbent in the atmosphere of the sun's radiation. A high temperature means generally a high saturation deficit, which allows of more intense insolation. Thus, temperature and solar radiation have in this experiment a correlation of 0·44, temperature and saturation deficit 0·71, and saturation deficit and insolation 0·66.

Similarly evaporation is correlated with insolation and sun temperature, although the atmometer which measured it is white, and does not register the component of air drying power due to radiant heat. The correlation is spurious—a reflection merely of the insolation-humidity correlation, which is itself accountable largely as an indirect effect of temperature acting on the humidity factor.

The correlations mentioned are "total" correlations; they are those obtained in nature, and may vary from one climatic region to another. For this reason, as well as in order to examine the "pure" effect of each factor on the dependent variable being considered, it is necessary to deal with the rather artificial concept of the "isolated" factor, the variations of which are imagined to be uncomplicated by corresponding variations of related factors.

When possible temperature effects on insolation are eliminated, the correlation of saturation deficit and insolation is little affected. But when the correlation of temperature and insolation is examined with the effects due to simultaneous related variations of saturation deficit removed, the apparent correlation disappears. The apparent correlation of evaporation and insolation, which gave a crude correlation of 0·61 for T_s and E, drops to 0·27 when the saturation deficit influence is removed.

Partial correlation of meteorological factors with oviposition.

The partial correlation of temperature and oviposition with saturation deficit eliminated is 0·46; when insolation is eliminated, it is 0·56. When both factors are eliminated, the second order partial correlation is 0·46. Thus, although insolation, increasing independently of temperature, depressed the oviposition response to the latter, the indirect effect of insolation changes related to those of temperature appears to be negligible. Saturation deficit was to some extent responsible for the effect credited to temperature, but the true effect of temperature, when this contribution is removed, remains relatively high.

The effect of air moisture was examined by using relative humidity values instead of saturation deficit. The intereffect is insignificant, the correlation dropping only to 0·55 from the crude 0·59. The evaporation rate, as a measure of air moisture, was also tried for possible partial responsibility for the apparent temperature effect. The partial correlation with evaporation eliminated is still 0·54. Thus air temperature is in itself responsible for practically all the oviposition results associated with it.

On the other hand, the partial correlations both of saturation deficit and of insolation, when temperature is eliminated, become insignificant :—

$$r_{YSD.T} = -0{\cdot}02 \text{ and } r_{YSR.T} = -0{\cdot}06$$

Elimination of saturation deficit also reduced the insolation correlation to insignificance (partial $r = -0{\cdot}07$).

The second order correlations, with two factors eliminated are :—

$$r_{YSD.SRT} = 0{\cdot}01\,; \quad r_{YSR.SDT} = -0{\cdot}05$$

The sun temperature, used as an alternative to insolation, fared equally badly. When saturation deficit was eliminated, the correlation became insignificant ($r=0{\cdot}1$).

As a second approach to the humidity relationship, the intereffects of the temperature in the sun and of evaporation rate on the saturation deficit correlation were tested. The second order partial correlation is :—

$$r_{YSD.T_sE} = 0{\cdot}21$$

which, though low, is significant. The reduction is principally due to the sun temperature factor, which when eliminated alone leaves an r value of 0·22.

Humidity expressed as relative humidity also ceases to have any significant correlation when temperature is eliminated, r becoming $-0{\cdot}01$. The evaporation value is no more significant by itself, $r_{YE.T}$ becoming 0·01.

The moisture content of the air would thus appear to have no significant effect in itself on oviposition, the considerable apparent correlation, 0·41, being due to associated temperature effects.

Wind.

An approximate estimate of the significance of wind may be obtained indirectly from the partial correlation of oviposition and evaporation, with the saturation deficit effect eliminated, since the atmometric index measures temperature, humidity, and wind.

$r_{YE.SD}=0{\cdot}014$, *i.e.* there is no significant correlation.

Wind would thus appear to have had no effect on oviposition, within the limits of this experiment. This finding is, however, much invalidated by the absence of any measure of the range of wind strengths encountered, and also by the crude grading of the evaporation scale, which possibly influenced the result that the total correlation for evaporation was only 0·27.

The various partial correlations are summarised in Table IX.

TABLE IX.

Corr.	Total r.	Eliminated	Partial r.	Corr.	Total r.	Eliminated	Partial r.
YT	0·586	SD	0·463	YT	0·586	RH	0·548
		SR	0·559			E	0·540
		SR and SD	0·461				
YSD	0·406	T	−0·021	YSD	0·406	T_s	0·217
		SR	0·357			E	0·314
		T and SR	0·010			T_s and E	0·208
YSR	0·218	T	−0·055	YT_s	0·367	SD	0·113
		SD	−0·070			E	0·264
		T and SD	−0·052				
YE	0·272	SD	0·014	YE	0·272	T	0·010
		T_s	0·065	YRH	−0·249	T	−0·013
		SD and T_s	−0·017				

Multiple Regression of Oviposition on Temperature, Humidity and Insolation.

The partial regression coefficients for the multiple regression equation are: $y-\overline{y}=0{\cdot}043\ (T-\overline{T})+0{\cdot}002\ (SD-\overline{SD})-0{\cdot}001\ (SR-\overline{SR})$, the partial regression of solar radiation being negative. The total regression of temperature, to four figures, is 0·0419, the partial 0·0428. As shown in Table X, however, the amount of the variance absorbed by the regression as fitted from the multiple equation is practically identical with that due to the simple regression on temperature when the other factors are neglected, the error variance for the latter being 0·07357 and for the multiple regression, with two more degrees of freedom absorbed, 0·07471. Thus, the partial regression line is no more descriptive a statistic of the data than is the simple regression.

TABLE X.

Source of variance	Degrees of freedom	Sum of squares	Mean square
		Total regression of T	
Total	133	14·8395	0·1116
Regression line	1	5·1278	
Residual	132	9·7117	0·07357
	Multiple	regression on T., S.D.	, and S.R.
Total	133	14·8395	0·1116
Regression function ...	3	5·1267	1·7089
Residual	130	9·7128	0·07471

Z for multiple regression 1·565. P<0·01.

From Table X the multiple correlation factor for oviposition with the three x factors can be calculated. It is 0·5878 as against 0·5856 for temperature, a quite insignificant improvement in the correlation, indicating the negligible extent to which oviposition depends on the combined variation of the three factors.

The simple regression of oviposition on temperature is illustrated in fig. 3. For this, the temperature intervals have been taken as half degrees. This, while not affecting the regression as expressed in 1 degree units, has improved the correlation, r being 0·595 as against 0·586. In comparison, the regression line is drawn for all temperatures above the lower critical limit of 58°F. With this selected data the correlation has decreased to 0·507 and the regression slope has increased from 0·042 to 0·056 for each degree.

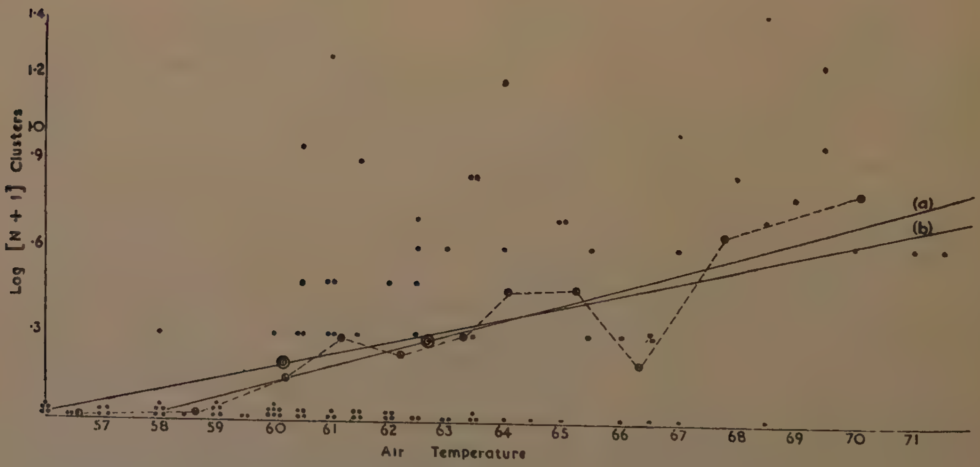

Fig. 3.—The correlation of oviposition (number of egg clusters) and mean air temperature for the hour. Tests at temperatures below 56° (omitted from the diagram), were all negative. The regression line (*a*) is for tests at temperatures of 58°F. and over, and (*b*) is for all tests. The group means are joined by the interrupted lines.

The regression is linear, for log oviposition and temperature, *i.e.* the oviposition increases in geometric ratio for arithmetic increase of temperature. The coefficient of 0·043 means an increase of 10·4 per cent. for 1 degree of temperature, or (since each successive value is 10·4 per cent. of the preceding value) a doubling for 7° increase of temperature. The curve for the true regression of oviposition on temperature is thus of the Compound Interest Law type; its cartesian equation will be :—

$$y+1=\bar{y}_1\, 1{\cdot}104^{x-\bar{x}}$$

(where $\bar{y}_1$ is the mean of the $y+1$ values, *i.e.* of the cluster values with unity added to each) or more simply $y+1=(\bar{y}+1)\, 1{\cdot}104^{x-\bar{x}}$.

Discussion.

It is clear that, within the range of temperatures covered by this experiment, temperature is in itself responsible for practically all of the oviposition variations which occurred. Humidity and insolation appeared to have little inherent influence, and the apparent relation of oviposition to these two factors is largely traceable to the insidious "undercover" activity of temperature.

The other factors are, however, seemingly not entirely devoid of effect. High insolation was found to reduce the oviposition for a given temperature, *i.e.* the two are antagonistic. The partial correlation of oviposition and insolation supports this finding; elimination of temperature or of saturation deficit leaves a correlation factor of —0·06 and —0·07 respectively. Though insignificant statistically, these values are high enough to make their negative sign suggestive.

Humidity, in this experiment, had no significant effect on the temperature-oviposition relationship. Humidity and insolation are nevertheless related, and this raises the possibility that low humidity may be responsible for some or all of the apparent insolation effect. This makes it worth while to examine the evidence for humidity, however slight it is.

(1) Oviposition was found to be associated with low humidity. The association is not statistically significant; if it exists, it is in any case probably a temperature effect.

(2) For a given temperature, high humidity had an adverse but insignificant effect on oviposition.

(3) For a given temperature, low evaporation, which is to some extent equivalent to high humidity, had an adverse, but insignificant, effect on oviposition.

(4) Evaporation had also a positive relationship to oviposition for a given insolation value, but this is probably a temperature effect.

(5) Although the partial correlation of oviposition and saturation deficit, with temperature eliminated, is extremely low and insignificant, the partial correlation of oviposition and temperature with saturation deficit eliminated, drops appreciably, indicating that some of the effect ascribed to temperature was due to high moisture deficit.

(6) The partial correlations of oviposition with R.H., S.D., and evaporation, temperature being eliminated, are so low that the mutually contradictory signs:

$$(r_{YRH.T} = -0{\cdot}01;\quad r_{YSD.T} = -0{\cdot}02)$$

are meaningless. The important point is that the relationship of oviposition and humidity, when isolated from temperature effects, is neutral. It may be argued that to maintain this neutrality in face of the insolation impress, *i.e.* to avoid being influenced by insolation, which would tend to vary simultaneously with it, humidity must have had a slight antagonistic, or compensating bias :

Decrease in humidity → insolation more intense, and so
→ increase in adverse effect.
→ Humidity decrease exercises favourable effect.
Resultant effect nil.

On balance, therefore, it would appear that, with further evidence, humidity may be found to have a significant effect on oviposition, increased humidity depressing the oviposition response to a given temperature.

Whether this be true or not, it seems reasonable to assume that the opposite is unlikely, *i.e.* humidity is not likely to discover a positive relationship with oviposition. The insolation effect must thus be a true effect of insolation ; it has possibly expressed itself in the teeth of a masking effect from humidity but has almost certainly not been accentuated by synchronous humidity effect.

The intensity of effect of insolation and humidity is, however, very low in comparison with the effect of temperature, and, broadly speaking, variations in oviposition, within the climatic limits of this experiment, can be ascribed to temperature changes, whether the variations are observed to be correlated directly with temperature, or with the air moisture index, which is directly related to it, or with the intensity of insolation, which is indirectly related through its dependence on air moisture.

The partial regressions of oviposition on humidity and insolation can be neglected as not significantly differing from zero regression. That is, the regression on temperature, whether taken as a total or partial regression, explains, in this experiment, all that portion of the variation in oviposition which can be mathematically accounted for. This amounts to about 35 per cent. of the total variability, and the remainder must be grouped as unexplained "error". This error is probably due to a variety of causes, of which the conventional error—of technique, recording, personal bias, etc.—is only one. Many of the others can be grouped under the heading, *changes in "availability" of flies.*

The contributory causes of such changes are many. The population density may fluctuate, from remote cause (*e.g.* earlier variations in strike incidence, so that the emergence rate is irregular) or from immediate cause, such as climatic impress ; the proportion of gravid females in the population is dependent on the inconstant supply of protein, *e.g.* as carrion, since a protein meal is necessary for maturation of ovaries ; the distribution of the population may be affected by variations of the total sheep stock on the farm, or by changes in their distribution from one field to another, and so on.

With regard to experimental error, a truer picture would almost certainly have been obtained by using more than one type of attractant, since little is known of the exact identity or identities of the odour operating in nature, and of any possible changes in its identity with changing meteorological conditions.

The possibility of errors of treatment of the data cannot, unfortunately, be excluded from consideration, but as a check on the possibility of gross arithmetical error the correlations were recalculated, using slightly different intervals of the x factors. It is hoped that all major errors of arithmetic have been detected.

The effect on the assessment methods of the abnormal distribution of log values for oviposition is unknown ; it is probably important, since the theory of both the correlation and regression formulae assumes normality of the distributions.

Summary.

The relationship of oviposition to weather, with a standard stimulus (4 per cent. ammonium carbonate), has been tested by a series of 134 exposures of a group of three sheep for one hour. 190 egg clusters were obtained, of which 169 were identified. With the exception of one cluster of *L. caesar* all were *L. sericata.*

There was a fairly close correlation (0·586) of oviposition with air-temperature. When the possible effect of other factors was eliminated, the correlation was little affected, showing that temperature was in itself responsible for the variations in oviposition. The lower critical limit of temperature was in the neighbourhood of 56°–58°F.

Solar radiation and oviposition had an apparent correlation, but this was found to be spurious, the effect being due to related air-moisture changes. At temperatures near the lower critical limit, insolation did not increase the oviposition rate, or lower its temperature threshold.

Humidity had an apparent inverse correlation with oviposition, but this also was an indirect effect, due to related temperature changes. Humidity showed a closer correlation when expressed as saturation deficiency than as relative humidity. Evaporation rate from a white atmometer sphere had only a low correlation with oviposition.

It is shown that the partial correlations of both saturation deficit and insolation with temperature effects eliminated, are insignificant, *i.e.* their apparent effects are due to related temperature changes. The multiple correlation coefficient for temperature humidity and insolation is practically identical with that for temperature.

Independent variations of insolation, *i.e.* variations not simultaneous with related temperature changes, had a significant effect on oviposition ; for a given temperature, a high intensity of insolation was less favourable than a low, and this appeared to be a true insolation effect, and not due to associated humidity changes.

Independent variations of humidity had a suggestive but statistically insignificant effect, humidity tending to be inversely related to oviposition.

The regression of log oviposition on temperature, within the limits of this experiment, *i.e.* at temperatures up to 72°F., was linear. The slope was not significantly affected by the insolation and humidity factors, the total coefficient being 0·0419, and the partial coefficient 0·0428. These coefficients are the increase of the log number of egg clusters (plus one) for 1 degree F. increase, *i.e.* the oviposition rate increased in geometric progression for unit increase of temperature, the value (N+1) for egg clusters doubling itself for 7° increase.

Acknowledgements.

I would like to put on record my indebtedness to my assistant K. W. Page, who was entirely responsible for the experimental results, including the meteorological records, and also for the rearing and identification of the cultures.

NOTES ON THE TACHINID PARASITES OF COTTON STAINERS (*DYSDERCUS* SPP., PYRRHOCORIDAE) IN SOUTH AFRICA.

By R. C. Rainey, A.R.C.S., B.Sc., Ph.D.
Empire Cotton Growing Corporation, Barberton, Transvaal.

E.M.N

Cotton stainers of the genus *Dysdercus*, vectors of the lint-staining fungus *Nematospora*, are major economic pests of cotton in South Africa. Tachinid parasites of *Dysdercus* were reported from Nigeria in 1923 by Pomeroy and Golding, and from Queensland in 1927 by Curran, but early searches for these or any other parasites of cotton stainers in South Africa gave negative results (Ullyett 1930). In 1931, however, Tachinid parasites of *Dysdercus* were recorded from Barberton, in the eastern Transvaal, though only 1·3 per cent. of a total of about 60,000 stainers examined over a period of three months were reported as parasitised (Parsons 1932). Stainer damage to cotton on the Experiment Station at Barberton in 1931 was the worst ever experienced, and the Tachinids appeared to be of little economic value. Identifications were not obtainable at this stage, but four species were considered to be present. Later work on *Dysdercus* in Uganda (Hargreaves & Taylor 1938) showed that parasitism by the Tachinid, *Bogosiella fasciata*, F., was at times very heavy, particularly on wild host-plants, and it was therefore suggested (Pearson 1939) that the subject might usefully be re-investigated at Barberton.

12 27
16 157
376

Stainer and Tachinid Populations in the Field.

In May and June 1940 about 1,900 adult and fifth-instar stainers, from routine weekly collections in a heavily infested field of well-grown late-planted Upland

TABLE I.

Populations of Dysdercus nigrofasciatus *and its Tachinid parasites on late cotton at Barberton, Transvaal.*

	1st collection 24th May			2nd collection 31st May			3rd collection 14th June		
	♂♂	♀♀	VV	♂♂	♀♀	VV	♂♂	♀♀	VV
No. of stainers per acre	9,400	5,100	3,700	11,400	5,800	13,700	11,800	9,300	16,600
Percentage parasitism : Total	25	26	15	35	34	3	24	22	No record
By *Alophora nasalis*	18	19	7	26	27	0	18	15	
By *Bogosiella fasciata*	2	2	7	2	1	2	2	2	

Notes :—

(i) Stainers collected on duplicate composite plant samples (Pearson, 1934), each comprising five 20-plant units of total area 1/72 acre, distributed over an area of 1½ acres within a 2-acre field of U.4 derivatives 920 and 6128, planted 13.xii.1939 and first picked 30.v.1940.

(ii) Differences in estimated stainer populations required for significance at 5 per cent. level (17th May–21st June inclusive) :—
♂♂ 3,300 per acre
♀♀ 2,100 „
VV 8,400 „

(iii) Analysis of numbers of adult stainers collected, and of total numbers of parasites on each occasion, gives highly significant differences between occasions in proportion of stainers parasitised ($\chi^2 = 18{\cdot}51$, $n = 2$ and $P < 0{\cdot}01$), but no significant difference between sexes of host in this respect ($\chi^2 = 0{\cdot}34$, $n = 3$ and $P > 0{\cdot}95$).

cotton, were transferred to the insectary to provide material for the identification of the parasites concerned, and to afford some indication of their incidence on cotton at the end of the season under local conditions. Eventually 339 Tachinids emerged, and identifications were subsequently obtained from the Imperial Institute of Entomology. The results for *Dysdercus nigrofasciatus*, Stål, representing over 99 per cent. of the stainers collected, are summarised in Table I, together with estimates of the corresponding stainer populations.

While great variation in the parasitism of stainers has been observed (Ullyett 1931, personal communication), both between and within seasons, it may be noted that Table I gives figures for percentages of parasitism on cotton at the end of the season which are considerably higher than corresponding figures recorded either in Uganda (Hargreaves & Taylor 1938), or nine years previously at Barberton (Parsons 1932), though lower than those found on wild host plants in Uganda. The sexes were parasitised to a similar degree; in Uganda *B. fasciata* was stated to attack a slightly higher percentage of female stainers than of males.

The proportions of parasitised stainers in the three collections differed significantly, to an extent which can scarcely be attributed to variations in adult Tachinid activity within the area sampled. On such an assumption the data for parasite numbers in successive collections, in conjunction with the insectary survival and emergence figures, give estimates of 47 per cent. of the parasitised stainers in the 2nd collection as having been infected between 25th and 31st May, and less than 1 per cent. of those in the 3rd collection as infected between 1st and 14th June. The absence of any significant difference between the 2nd and 3rd collections in the period between host collection and parasite emergence (the respective mean periods were 37 and 35 days) is inconsistent with such an abrupt decline in Tachinid activity in early June, and the meteorological records also afford no indication of any appreciable change in the weather over this period.

Most of the increase in the parasite population between the 1st and 2nd collections may reasonably be attributed to a relatively high level of parasitism in immigrant stainers. The birth and death-rates of adult stainers, estimated from the insectary observations on corresponding nymphs and adults, suggest a net excess of immigrants over emigrants of about 40 per cent. of the adult population per week over this period. It has, furthermore, been shown that emigration of stainers is almost always in progress (Pearson 1939), even from host plants in a favourable condition (when it is offset by immigration on an equal or greater scale), so that the actual extent of immigration, particularly prior to the first picking, is likely to have been greater than this figure suggests.

Between the 2nd and 3rd collections the estimated net immigration rate was considerably lower, about 16 per cent. of the adult population per week, and the first adults bred up in this particular crop were also beginning to make their appearance. The observed decline in the parasite population in the field over this period indicates that the combined numbers of parasitised immigrant stainers, and of stainers parasitised within the sample area during these two weeks, failed to make good the corresponding losses of parasite larvae by emigration of hosts, emergence, etc.

The small proportion of nymphs parasitised by *Alophora nasalis*, in comparison with the percentage of adults attacked by this species, may indicate either a strong preference for adult hosts, or merely a relative scarcity of adult *A. nasalis* in this particular field; the immigration figures and the lengthy larval development of the parasite suggest that most of the parasitised adults recorded are likely to have been attacked before their arrival in the sample area.

The predominant species of Tachinid, *A. nasalis*, Bezzi, has been recorded under various synonyms (*Allophora nigeriensis*, Villen., *Hyalomyia cuthbertsoni*, Curr.) from Cape Province to Kenya and Nigeria (Cuthbertson & Munro 1941; van Emden

1945), including Southern Rhodesia (Gatooma), Northern Rhodesia (Mazabuka) and the Congo (Boma). A series of specimens illustrating the range of variability of the present material has been retained at the British Museum (Natural History). *Bogosiella fasciata*, F., which was present in small numbers, is the predominant species in Uganda, particularly in the wetter parts (Hargreaves & Taylor 1935), and ranges from Sierra Leone to Nyasaland and from the southern Sudan to Pondoland (van Emden 1945). It appears most frequently in regions of high rainfall, most of the localities from which it has been recorded receiving 40–80 in. per annum, as compared with 20–40 in. for *A. nasalis*; Barberton's annual rainfall averages 28 in. Laboratory observations in Uganda showed that the full-grown larva of *B. fasciata*, after emergence from its host, burrows into the soil to form the puparium, and that considerable moisture is necessary for successful pupal development (Hargreaves & Taylor 1938), which may be a limiting factor in the distribution of the species. In the course of the earlier observations at Barberton, on material subsequently identified as *A. nasalis* (van Emden 1945), it was noted that this species pupated always on the surface of the soil (Ullyett, personal communication), and in the present investigation satisfactory pupal development was obtained without any attempt to keep the puparia moist.

Tachinids from *D. nigrofasciatus* collected on *Hibiscus calyphyllus*, Cav., the most important local wild host-plant of this species, along the Helahela and Lamfomo spruits near Bremersdorp in Swaziland, were also identified at the Imperial Institute of Entomology as *A. nasalis* and *B. fasciata*.

A few specimens of other species of *Dysdercus* were collected with the *D. nigrofasciatus* recorded in Table I. Of two males, two females and two fifth-instar nymphs of *D. fasciatus*, Sign., one adult of each sex and both nymphs proved to be parasitised by *A. nasalis*, while of two *D. intermedius*, Dist., of each sex, one male was parasitised by the same Tachinid. *A. nasalis* has been recorded from *D. fasciatus*, *D. intermedius*, *D. nigrofasciatus* and *D. superstitiosus*, F., and from the Pentatomid, *Agonoscelis versicolor*, F. (Cuthbertson & Munro 1941, van Emden 1945). *B. fasciata* has been recorded from *D. cardinalis*, Gerst., *D. fasciatus*, *D. nigrofasciatus*, *D. superstitiosus* and *D. ugandanus*, Schout. (Hargreaves & Taylor 1938, van Emden 1945).

The Development of *Alophora nasalis* **and** *Bogosiella fasciata*.

After being brought in from the field, the stainers were kept on a diet of cotton-seed in an unheated open-air insectary, at temperatures which averaged 57–60°F. during June and July.

TABLE II.

Mean period between date of collection of D. nigrofasciatus *and emergence of Tachinid larva.*

Nature of host	*Alophora nasalis*	*Bogosiella fasciata*
Male stainers	34 days (179)	14 days (17)
Female stainers	41 days (108)	26 days (10)
5th instar nymphs ...	74 days (7)	60 days (13)

Differences between male and female hosts are significant (for *A. nasalis* $P<0{\cdot}01$; for *B. fasciata* $P<0{\cdot}05$); those between collections are not.

Number of parasites recorded in brackets.

Table II shows that under these conditions the development of the Tachinid larvae, notably *A. nasalis*, within their hosts was a lengthy process, particularly since infection occurred an unknown length of time prior to the date of collection from which the figures of Table II are reckoned. At Kampala, with temperatures probably 10° or more higher, the larval development of *B. fasciata* was found to take 13–16 days in adult hosts, and about 10 days longer in stainer nymphs (Hargreaves & Taylor 1938). In the present material, emergences of *A. nasalis* were frequent up to 9–10 weeks; the last parasites emerged from male stainers during the 13th week after collection, and from female stainers during the 12th week, by which time 66 per cent. and 70 per cent. of the respective corresponding unparasitised stainers had also died. The mean larval period must thus have been of the order of 2–3 months. The development of the parasites within nymphal hosts was even more protracted, with emergences of both species recorded up to the 15th week after collection; several parasitised nymphs completed their final moult successfully before the emergence of the Tachinid larva. Parasitised stainers were thus surprisingly long-lived, and a few individuals even survived the emergence of the parasite, as has also been observed elsewhere (Ballard & Evans 1928; Hargreaves & Taylor 1938; Pomeroy & Golding 1923). The protracted larval development of *A. nasalis* may be a factor of some importance in the survival of this species during the cool dry season, from July onwards, when *D. nigrofasciatus* is represented by small populations of widely dispersed adults, which re-appear on perennial Malvaceae when growth is resumed at the break of the rains in November (Pearson 1939).

There was no significant difference between the three collections of adult stainers in the length of time from the date of collection of the hosts until the emergence of the parasites, but, while percentage parasitism was independent of sex of host, both species of Tachinid emerged earlier from male than from female stainers in all three collections, a difference which was statistically significant. It may therefore be suggested that the duration of larval development is greater within female than within male hosts. Such a difference may perhaps be associated with the fact that in both species individuals from female stainers were noticeably larger than those from males. In *A. nasalis*, for example, the mean head-width, including the eyes, was 2·40 mms. in flies from male hosts, as compared with 3·15 mms. in flies from females, the difference being highly significant ($P < 0{\cdot}01$).

The mean pupal period at a constant temperature of 80°F. was 15·3 days in *A. nasalis*, with a standard deviation of 0·35 days, and $14{\cdot}0 \pm 0{\cdot}65$ days in *B. fasciata*, for which species pupal periods of 12·4 and 13–17 days have been recorded at Ibadan (Nigeria) and Kampala respectively (Hargreaves & Taylor 1938, Pomeroy & Golding 1923).

The Economic Significance of the Parasites.

Estimates of the economic significance of the Tachinid parasites of *Dysdercus* have varied considerably in different territories. Thus *Acaulona peruviana*, Tns., and *Hyalomyia chilensis*, Macq., reared from *Dysdercus ruficollis*, L., in Peru, were considered of sufficient economic value to warrant their introduction into the island of Porto Rico in 1942 for the control of *D. andreae*, L.; species of *A. peruviana* were recovered in one locality the year after release (Bartlett 1943). In Uganda it was stated that in 1936 the control of stainers on wild host plants could apparently be left to *B. fasciata*, but that on seed-dumps it could not (Hargreaves & Taylor 1938). In Nigeria the relatively slight effects of parasitism by Tachinid larvae on the longevity and even the fecundity of *D. superstitiosus*, F., were regarded as indicating that the parasites do not constitute an effective natural control and are of no real economic importance (Pomeroy & Golding 1923).

The present observations, while indicating higher levels of parasitism than had previously been recorded either locally or in Uganda, have also demonstrated the surprising longevity of parasitised stainers. Furthermore, the heavy stainer attack of 1931 afforded no indication of effective parasite activity. So far there is in fact no local evidence of any serious effects on stainer numbers attributable to the Tachinids, whose economic value appears correspondingly slight, though further observations, particularly of parasite activity on wild host-plants, might still be of considerable interest. Both the Tachinids concerned are widely distributed indigenous species, and their present relatively amicable relationship with their hosts may perhaps have developed in the course of a prolonged association.

The results obtained have also some bearing on the interpretation of stainer marking experiments carried out on *Hibiscus calyphyllus* near Barberton. These experiments indicated that the time of maximum stainer breeding was also the time of most active dispersal, but it was suggested that a seasonal rise in parasitism might provide an alternative explanation of the small proportion of marked stainers recaptured over this period (Pearson 1939). However, even if parasitism were to attain a high level over the period concerned, the observed longevity of parasitised stainers (though admittedly recorded under winter conditions) indicates that a rise in the death rate sufficient to account for the decline in the recapture figures would still be unlikely.

Summary.

(1) Up to 35 per cent. of adult *Dysdercus nigrofasciatus* from a heavily infested late cotton crop at Barberton were found to be parasitised by Tachinid larvae at the end of the season.

(2) The predominant Tachinid species was *Alophora nasalis*; *Bogosiella fasciata* was present in smaller numbers, and appears to be more common in regions of higher rainfall.

(3) The development of the Tachinid larva within the stainer was a protracted process under local winter conditions, the larval period of *A. nasalis* being of the order of 2–3 months, and parasitised stainers were correspondingly long-lived.

(4) The possible economic significance of the parasites is discussed.

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DDT RESIDUAL FILMS.

I.—THE PERSISTENCE AND TOXICITY OF DEPOSITS FROM KEROSENE SOLUTIONS ON WALL-BOARD.

By E. A. Parkin & A. A. Green.
Pest Infestation Laboratory, Slough, Bucks.

Introduction.

An outstanding property of DDT is its relative stability under practical conditions, a property which is of especial value in the control of such insects as flies, mosquitos, bed-bugs and lice. The formation of persistent insecticidal films of DDT upon various surfaces has been reported by numerous workers and the use of such films for control of insects of medical importance is now being rapidly developed in many countries. Work on the use of DDT in the control of the house-fly, *Musca domestica*, L., has been in progress at the Pest Infestation Laboratory since 1943 and, as an extension of an investigation into the use of DDT in fly sprays (Parkin & Green, 1945a), attention was turned, at the request of the War Office and the Insecticide Development Panel, to the question of the formation and persistence of residual films of DDT on different types of solid surfaces.

The investigation was divided initially into the following sections :—

1. The formation and persistence of residual films from a kerosene solution of DDT on a convenient surface, including the effects of concentration, deposit, etc.

2. The formation and persistence of DDT films on different surfaces.

3. A comparison of different methods of application of DDT to surfaces, *e.g.* in oil solution, emulsions, dispersible powders, distempers, paints, etc.

This paper reports the results so far obtained in the first section of the investigation. The house-fly has been used as the test insect. A proprietary brand of wall-board was chosen as a suitable substrate, because it was a material frequently encountered in practice and a stock of sheets of reasonably uniform manufacture was obtainable. Glass was found to be unsuitable, since flies affected by DDT generally vomit and, if they fall on their backs, become stuck to the glass : furthermore, the use of such a non-absorbent surface greatly restricts the range of deposits which can be tested.

Experimental Technique.

The wall-board, an absorptive material made from compressed paper and about 0·19 in. thick, was sawn into pieces 6 in. square. Each square was sprayed on one side with insecticide by means of a scent sprayer, fitted with a small glass liquid-reservoir and worked by compressed air at 3 lb./sq. in. Using this simple apparatus, a reasonably even distribution of the relatively coarse spray could be obtained by holding the sprayer about 2 in. away from and traversing backwards and forwards across the surface of the square. Very little spray was lost by rebound, the chief loss occurring during treatment of the periphery of the board. Several weighed boards were sprayed with different amounts of Pool burning oil and immediate reweighing showed that between 80·5 and 81·7 per cent. of the liquid measured into the reservoir of the sprayer had been deposited on the boards. In subsequent calculation of dosages it was therefore assumed that 80 per cent. of the spray adhered to the target. Four samples were sprayed with each concentration, deposit, etc., of DDT solution. All treated and untreated control squares were stored in a room maintained at 27·5°C. and 60 per cent. R.H. At least 4 days elapsed before their first exposure to flies, by which time the odour of the solvent was no longer detectable.

The DDT used in these tests was a technical grade product, 77 per cent. pure, and the concentrations of solutions and weights of deposits are given in terms of the technical product. Solutions were made in Pool burning oil, the British war-time grade of kerosene.

The test insects were adult house-flies, *Musca domestica*, L., reared on an artificial diet at 27·5°C. and 60 per cent. R.H. The flies were fed on a 1 : 1 mixture of milk and water and were 4–6 days old when used. Immediately before exposure on the squares, flies, in batches of about one hundred in muslin-covered wire cages, were inactivated by chilling at 2°C. for 15 min. The flies were tipped on to a cold enamelled tray and quickly transferred, by gently holding a leg or wing with forceps, to the samples of wall-board. Ten flies were placed on each sample and covered by an inverted petri dish, 10 cm. in diameter. The flies appeared to have recovered their normal activity within 3–4 min. of being replaced at 27·5°C. and 60 per cent. R.H., under which conditions all subsequent observations were made. In some experiments in which the exposure period exceeded 4 hours a small piece of cork was fixed in the centre of each sample and carried a wad of cotton wool soaked in water. The flies usually visited the cotton wool for a drink only after confinement for about 5 hours and showed no tendency to congregate on the cork or the wad in preference to the wall-board.

The number of flies " knocked down " on each square was recorded after various periods of exposure. A fly was considered to be knocked down if it had turned on to its back and was unable to regain the normal position, or if it remained on its feet but had lost the power of co-ordinated movement. The use of knock-down as a measure of toxicity had the advantage of avoiding the need for transference of flies, after an arbitrarily fixed period of exposure, back to muslin cages for later observation of mortality. The train of toxic symptoms was similar to that described by Parkin, and Green (1945a) for sprays of DDT in kerosene. It must be borne in mind, however, that knock-down as described for DDT is very different physiologically from the reversible knock-down caused by an insecticide like pyrethrum.

Some Factors affecting Film Formation.

Preliminary experiments of relatively short duration were made in order to determine suitable levels of concentrations and deposits for the main experiment, in which it was planned to measure the toxicity of the films at intervals for a year or more. In addition, *ad hoc* experiments were carried out to observe the formation and fate of DDT crystals on the wall-board. Over 2,000 observations were recorded in the main experiment, and drastic condensation has been necessary in presenting the table of results. In order to show the type of replication obtained with the technique employed, the results of one of the preliminary tests on the effect of deposit are given in full.

Effect of concentration.

Three lots of four boards were sprayed with 10, 2 or 0·2 per cent. w./v. DDT in Pool burning oil so that each board received a deposit of 80 mg. DDT/sq. ft.* Flies were exposed on the boards 4 days and 4 weeks after spraying. The results are summarised in Table I.

The residual film formed from a small deposit of 10 per cent. DDT in kerosene was markedly more effective in speed of knock-down than the same weight of DDT applied in a larger volume of the solvent. Moreover, the films formed by the 2 and 0·2 per cent. sprays lost toxicity during the 4 weeks storage period, whereas those formed by the 10 per cent. spray increased in toxicity. The toxicity of a residual film of DDT on wall-board is therefore dependent on the concentration of the solution

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* 1 mg./sq. ft.=10·76 mg./sq. m.

TABLE I.

The toxicity to house-flies of films of 80 *mg. DDT/sq. ft. applied to wall-board from solutions of different concentrations.*

Concentration DDT w./v.	Exposed 4 days after spraying				Exposed 4 weeks after spraying			
	Mean knock-down (per cent.) after				Mean knock-down (per cent.) after			
	1 hr.	2 hr.	3 hr.	4 hr.	1 hr.	2 hr.	3 hr.	4 hr.
10 per cent. ...	30	57·5	100	—	42·5	92·5	100	—
2 ,, ...	5	17·5	52·5	77·5	0	0	10	30
0·2 ,, ...	0	0	0	12·5	0	0	2·5	7·5
Controls	0	0	0	0	0	0	0	0

used in making the application. In contrast to this result, Gahan & others (1945) found that, for equal deposits of DDT, toxicity to mosquitos was independent of concentration for kerosene solutions containing 1–4 per cent. DDT applied to wood or canvas.

Effect of deposit.

Deposits of 40, 80, 160 and 320 mg. DDT/sq. ft. were applied to samples of wall-board using a 5 per cent. solution. Flies were exposed on the boards 4 days and 4 weeks after spraying. Four replicates were made of each treatment and the results are set out in detail in Table II as a typical example of the extent of variation encountered during the investigation.

TABLE II.

The toxicity to house-flies of different deposits of DDT applied to wall-board from a 5 *per cent. solution.*

Deposit mg. DDT per sq. ft.	Board No.	Exposed 4 days after spraying				Exposed 4 weeks after spraying			
		Number K-D after				Number K-D after			
		½ hr.	1 hr.	2 hr.	3 hr.	½ hr.	1 hr.	2 hr.	3 hr.
40	1	0	3	9	10	3	7	10	10
	2	0	1	9	10	2	8	10	10
	3	0	4	10	10	4	8	10	10
	4	0	2	7	10	2	6	8	10
Mean K-D (%) ...		0	25	87·5	100	27·5	72·5	95	100
80	5	0	5	10	10	7	8	10	—
	6	0	5	10	10	8	9	10	—
	7	0	4	10	10	6	7	10	—
	8	0	4	9	10	8	9	10	—
Mean K-D (%) ...		0	45	97·5	100	72·5	82·5	100	—
160	9	4	7	10	—	8	9	10	—
	10	5	9	10	—	8	10	10	—
	11	5	8	10	—	8	9	10	—
	12	4	7	10	—	8	9	10	—
Mean K-D (%) ...		45	77·5	100	—	80	92·5	100	—
320	13	7	10	10	—	10	10	—	—
	14	8	10	10	—	10	10	—	—
	15	7	10	10	—	9	10	—	—
	16	6	9	10	—	8	10	—	—
Mean K-D (%) ...		70	97·5	100	—	92·5	100	—	—
Controls ...	...	...	0	0	0	0	0	0	0

Table II shows that, with a constant concentration of 5 per cent. DDT in Pool burning oil, an increase in the amount deposited on wall-board led to an increase in the toxicity of the residual film, at least within the limits investigated. Furthermore, all films increased markedly in toxicity between the first and second exposure to flies, as happened with the concentration of 10 per cent. in the previous experiment (see Table I).

The experiment was repeated but with a 2·5 per cent. solution of DDT and the resulting data are summarised in Table III.

TABLE III.

The toxicity to house-flies of different deposits of DDT applied to wall-board from a 2·5 per cent. solution.

Deposit mg. DDT per sq. ft.	Exposed 4 days after spraying					Exposed 4 weeks after spraying				
	Mean knock-down (per cent.) after					Mean knock-down (per cent.) after				
	½ hr.	1 hr.	2 hr.	3 hr.	4 hr.	½ hr.	1 hr.	2 hr.	3 hr.	4 hr.
40	0	0	10	27·5	55	0	0	0	2·5	20
80	0	0	22·5	65	100	70	87·5	100	—	—
160	0	2·5	52·5	95	100	85	97·5	100	—	—
320	2·5	20	87·5	100	—	95	100	—	—	—
Controls	0	0	0	0	0	0	0	0	0	0

The data in Table III confirm those of Table II in showing that, with a given concentration of DDT in Pool burning oil, the toxicity of the residual film increases as the amount of insecticide applied increases. Lindquist and others (1944) showed that the speed of knock-down of house-flies increased as the deposit of DDT on wood increased from 10 to 200 mg./sq. ft. On the other hand, Fay and others (1945) recorded little difference in the mortality of adult *Anopheles quadrimaculatus*, Say, exposed to deposits of 50 to 300 mg. DDT/sq. ft.

The rise in toxicity after the first period of exposure is again evident, except with the lowest deposit : a similar deposit applied from a 5 per cent. solution showed a rise (Table II). Finally, all deposits from 40 to 320 mg. DDT/sq. ft. applied from a 2·5 per cent. solution were less toxic at the first exposure than corresponding deposits from a 5 per cent. solution, but the pairs of deposits from 80 to 320 mg. DDT/sq. ft. applied from the two concentrations were about equally rapid in action at the second exposure.

The results of the three experiments so far described led to the following conclusions :—

1. The initial toxicity of a film of DDT on wall-board, measured by speed of knock-down of house-flies, is governed by the combination of deposit and concentration of the solution used—heavy deposit and high concentration favour rapid toxic action.

2. If some function of concentration and deposit has a value above a certain minimum, a film of residual DDT on wall-board may be expected to increase in toxicity after the first exposure to flies.

Effect of method of application.

Groups of four squares of wall-board were sprayed with 80 mg. DDT/sq. ft. in three different ways. At the start of the experiment, one group received one application of 0·5 ml. of 5 per cent. solution ; a second group received one application of

2 ml. of 1·25 per cent. solution ; and the third group an application of 0·5 ml. of 1·25 per cent. solution. The third group received three subsequent applications of 0·5 ml. of 1·25 per cent. solution at intervals of 2 days. The squares were exposed to flies 6 days after completion of the treatment of the third group, with the results shown in Table IV.

TABLE IV.

The toxicity to house-flies of 80 *mg. DDT/sq. ft. applied in different ways to wall-board. Tested* 6 *days after treatment.*

Treatment	Mean knock-down (per cent.) after			
	1 hr.	1½ hr.	2 hr.	2½ hr.
0·5 ml. of 5 per cent. DDT	30	55	55	75
2 ml. of 1·25 per cent. DDT	0	2·5	5	15
4×0·5 ml. of 1·25 per cent. DDT ...	0	5	25	32·5
Controls	0	0	0	0

Table IV shows that four applications of 0·5 ml. of 1·25 per cent. DDT in Pool burning oil gave a residual film slightly more toxic than a single treatment with 2 ml. of the solution, whereas one application of 0·5 ml. of 5 per cent. DDT gave a residual film much more effective than either of the equivalent treatments with the more dilute solution. It was therefore concluded that the best use of a given quantity of DDT would be made by applying it as a small dose of a concentrated spray. A 5 per cent. solution of DDT in Pool burning oil is about the highest concentration which can conveniently be used in practice, because of limitations of solubility, unless an accessory solvent is incorporated.

Penetration of DDT solution into wall-board.

In order to test whether DDT would be carried into the interior of wall-board if relatively large amounts of solution were used, boards were sprayed with a 5 per cent. solution at the rate of 80, 160, 320 and 1,280 mg. DDT/sq. ft. of treated surface. After spraying, the boards were left for 4 days at 27·5°C. and 60 per cent. R.H. and then split into laminae for exposure to flies. The results are summarised in Table V.

TABLE V.

Depth of penetration of 5 *per cent. solution of DDT into wall-board.*

Deposit mg. DDT/sq. ft.	Approx. depth from surface in.	Mean knock-down (per cent.) after	
		2 hours	3 hours
80	0	85	100
	0·04	0	15
	0·08	0	0
160	0	100	100
	0·04	5	35
	0·08	0	0
320	0	100	100
	0·04	30	70
	0·08	0	0
1280	0·10	100	100
	0·13		65
	0·16		8
	0·19		0

The exposure at 0·19 in. from the treated surface represented exposure of flies on the reverse side of the board. The data show that, as the deposit of spray increased, some of the DDT was carried by the solvent more and more deeply into the board, where it would be no longer available for killing insects alighting on the treated surface.

Crystallisation of residual films.

It has already been shown that, with certain combinations of concentration and deposit, the residual film left by a spray of DDT in Pool burning oil increased markedly in toxicity to flies between the 4 day and 4 week exposures. In the interim between the two exposures, those boards on which the film had become more effective were observed to have developed a lighter-coloured central area, corresponding with the area of the petri dish under which the flies had been confined. Examination of the surface of the boards under a binocular microscope (×30) with reflected illumination revealed that the lighter coloured area was thickly carpeted with crystals and that relatively few crystals occurred on the remainder of the surface. Sprayed samples not exposed to flies showed no development of a carpet of crystals during 5 weeks of observation and, when eventually exposed, exhibited a toxicity no greater than that of samples used 4 days after spraying.

Experiments with DDT solutions allowed to crystallise on glass plates proved that DDT has a great tendency to supersaturate and that this tendency is greater with crude than with pure DDT, and greater with Pool burning oil than with acetone as the solvent. The supersaturated solution formed highly viscous gummy droplets which crystallised on mechanical disturbance, *e.g.* with a needle or camel hair brush.

No more than very sparsely distributed single crystals or small bunches of crystals could be seen on treated wall-board before the first exposure to flies and it was therefore assumed that the DDT was present almost entirely in the form of the very viscous supersaturated deposit, which was markedly less toxic than crystalline DDT. Movement of the confined flies on the treated surface was sufficient mechanical agitation to induce general crystallisation and, as it were, to release the latent potential toxicity of the DDT residual film (Parkin & Green, 1945b). Visible signs of crystallisation could be detected 2 days after exposure to flies. The crystal carpet varied in density according to the deposit of DDT applied and the concentration of the original solution. Early exposure to flies after treatment caused the crystals to be shorter and more numerous than when exposure was delayed. The two types of crystalline films, however, were equally toxic.

The appearance of the crystal carpet also depended upon the type of stimulation applied to the supersaturated layer, for brushing or stippling the treated surface of wall-board with a soft paint brush gave rise to fewer but longer crystals, resembling those occurring in the annulus rubbed by the petri dish. On subsequent exposure to flies, the larger crystals induced by brushing were found to be as toxic as the smaller more numerous crystals formed by movement of flies. Barnes (1946) has reported indications that large crystals of DDT were less toxic to *Cimex lectularius*, L., than very small crystals.

Boards on which a good growth of crystals developed after exposure to flies, brushing, or stippling with a brush, were vigorously rubbed with a clean dry cloth, a treatment designed to emulate a severe cleaning process to which surfaces might be exposed in practice. Exposure to flies immediately after cleaning, when no crystals could be detected under the conditions of examination, showed that the DDT films had suffered a considerable reduction in toxicity, but, after storage for a further 10 days at 27·5°C. and 60 per cent. R.H., the cleaned boards had regained their toxicity, although crystal formation was still not detected. It must be assumed that the rubbing had removed many of the DDT crystals and had reduced the remainder to a size too small to be observed by the method used. Regeneration of the crystal layer had presumably started within the 10-day interval between the two exposures to flies. Regeneration of DDT crystals has also been reported by West and Campbell (1946) after rubbing over the surface of a coating of coumarone-resin varnish incorporating DDT.

Some samples bearing a thick carpet of DDT crystals were exposed to flies twice on the same day without any marked reduction in toxicity at the second exposure. Microscopic examination showed that relatively few crystals remained visible, and these only as broken stumps. Ten days later a new growth of minute crystals was apparent.

The observed increase in toxicity following the first exposure to flies may therefore be ascribed to the formation of crystalline DDT on the surface by mechanical agitation of the supersaturated solution of DDT which remains when most of the solvent has evaporated. The partial regeneration of the crystal layer after "removal" by rubbing or by exposure to flies is probably due to slow movement towards the surface of some of the DDT carried a short distance into the substance of the sample by the carrier and possibly held in the supersaturated state.

Microscopic examination of flies killed by exposure to these crystalline DDT films revealed broken crystals on all flies. The number of crystals on the flies increased as the deposit on the board increased over the range investigated (80–320 mg. DDT/sq. ft.). The broken crystals adhered to the body and to the setae and were most numerous on the ventral surface of the abdomen distally, on the wings, on the compound eyes, and on the tarsi.

Persistence of Residual Films.

On the basis of the results of the preliminary experiment, a major experiment was planned to investigate the persistence of toxicity of residual films of DDT on wall-board. Squares of wall-board were treated on 6th Nov., 1944, with 40, 80, 160 and 320 mg. DDT/sq. ft. from solutions containing 1·25, 2·5 and 5 per cent. DDT in Pool burning oil. For the whole of the experimental period, the samples were kept at 27·5°C. and 70 per cent. R.H. and, except during exposure to flies, were in the dark. Each combination of concentration and deposit was replicated four times and during exposures ten flies were confined on each sample. The knock-down was recorded after ½, 1, 2, 3 and 4 hours and the mean knock-down for each treatment determined for each exposure period. The samples were first exposed to house-flies 4 days after application of the DDT solutions and subsequently when the films were 4, 8, 12, 20, 38, 52 and 78 weeks old. At least 97·5 per cent. of flies kept on unsprayed boards during each exposure were classed as normal at the end of 4 hours. As a check on the general level of resistance of the fly stock, samples treated 4 days previously with 80 mg. DDT/sq. ft. from a 5 per cent. solution were exposed on each occasion, with the results shown in Table VI.

TABLE VI.

Variation in resistance to DDT (80 mg./sq. ft. on wall-board) of the fly stock during the experimental period.

Interval after start of experiment	Mean knock-down (per cent.) after				
	½ hr.	1 hr.	2 hr.	3 hr.	4 hr.
4 days	5	45	85	100	—
4 weeks	5	52·5	81·5	100	—
8 "	5	47·5	65	97·5	100
12 "	2·5	55	97·5	100	—
20 "	0	52·5	65	87·5	100
38 "	0	10	60	95	100
52 "	2·5	20	75	100	—
78 "	0	20	70	92·5	100

TABLE VII.

Persistence of toxicity of DDT films on wall-board, as measured by the mean percentage knock-down of house-flies, after different periods of exposure.

Age of film	Deposit mg. DDT/sq. ft.	5 per cent. DDT solution					2·5 per cent. DDT solution					1·25 per cent. DDT solution				
		Mean knock-down (per cent.) after														
		½ hr.	1 hr.	2 hr.	3 hr.	4 hr.	½ hr.	1 hr.	2 hr.	3 hr.	4 hr.	½ hr.	1 hr.	2 hr.	3 hr.	4 hr.
4 days	40	0	0	12·5	37·5	77·5	0	0	10·0	30·0	72·5	0	0	0	10·0	42·5
	80	5·0	45·0	85·0	100	—	0	0	30·0	90·0	100	0	0	2·5	22·5	67·5
	160	25·0	70·0	100	—	—	7·5	37·5	90·0	100	—	0	5·0	47·5	82·5	95·0
	320	52·5	80·0	100	—	—	32·5	62·5	100	—	—	5·0	52·5	92·5	100	—
4 weeks	40	60·0	77·5	100	—	—	0	0	10·0	30·0	52·5	0	0	2·5	5·0	22·5
	80	82·5	87·5	100	—	—	80·0	90·0	100	—	—	0	0	5·0	25·0	55·0
	160	92·5	95·0	100	—	—	85·0	90·0	100	—	—	90·0	97·5	100	—	—
	320	92·5	100	—	—	—	90·0	95·0	100	—	—	80·0	92·5	100	—	—
8 weeks	40	0	0	12·5	45·0	77·5	0	0	0	5·0	12·5	0	0	0	0	5·0
	80	67·5	80·0	100	—	—	30·0	70·0	97·5	100	—	0	0	0	2·5	17·5
	160	80·0	87·5	100	—	—	80·0	85·0	100	—	—	70·0	72·5	95·0	100	—
	320	90·0	95·0	100	—	—	82·5	87·5	100	—	—	80·0	85·0	100	—	—
12 weeks	40	0	0	0	7·5	22·5	0	0	0	5·0	12·5	0	0	0	7·5	10·0
	80	55·0	67·5	100	—	—	15·0	60·0	97·5	100	—	0	0	0	7·5	17·5
	160	80·0	92·5	100	—	—	62·5	82·5	100	—	—	27·5	62·5	100	—	—
	320	87·5	95·0	100	—	—	80·0	90·0	100	—	—	72·5	87·5	100	—	—

TABLE VII—*Continued.*

Persistence of toxicity of DDT films on wall-board, as measured by the mean percentage knock-down of house-flies, after different periods of exposure.

Age of film	Deposit mg. DDT/sq. ft.	5 per cent. DDT solution					2·5 per cent. DDT solution					1·25 per cent. DDT solution				
		Mean knock-down (per cent.) after														
		½ hr.	1 hr.	2 hr.	3 hr.	4 hr.	½ hr.	1 hr.	2 hr.	3 hr.	4 hr.	½ hr.	1 hr.	2 hr.	3 hr.	4 hr.
20 weeks	40	0	10·0	15·0	20·0	32·5	0	0	0	0	10·0	0	0	0	0	7·5
	80	55·0	82·5	100	—	—	0	45·0	77·5	87·5	100	0	0	0	7·5	20·0
	160	92·5	97·5	100	—	—	90·0	97·5	100	—	—	70·0	85·0	100	—	—
	320	97·5	97·5	100	—	—	90·0	97·5	100	—	—	90·0	97·5	100	—	—
38 weeks	40	0	0	0	2·5	7·5	0	0	0	0	0	0	0	0	0	0
	80	5·0	17·5	57·5	90·0	97·5	0	0	10·0	27·5	72·5	0	0	0	5·0	10·0
	160	92·5	95·0	100	—	—	77·5	87·5	100	—	—	2·5	32·5	75·0	92·5	100
	320	95·0	97·5	100	—	—	90·0	95·0	100	—	—	92·5	95·0	100	—	—
52 weeks	40	0	0	0	5·0	7·5	0	0	0	0	0	0	0	0	0	0
	80	0	0	0	22·5	42·5	0	0	0	0	22·5	0	0	0	0	0
	160	55·0	87·5	100	—	—	57·5	77·5	100	—	—	0	0	7·5	22·5	45·0
	320	95·0	97·5	100	—	—	87·5	92·5	100	—	—	80·0	85·0	100	—	—
78 weeks	40	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	80	0	0	0	0	7·5	0	0	0	0	7·5	0	0	0	0	0
	160	10·0	70·0	92·5	100	—	5·0	27·5	62·5	80·0	100	0	0	0	7·5	32·5
	320	92·5	95·0	100	—	—	85·0	90·0	100	—	—	57·5	85·0	100	—	—

During the first 20 weeks of the test the resistance of the stock remained reasonably constant.

After this a change occurred whereby the susceptible flies in the population became more resistant to DDT without any corresponding increase in resistance of the least susceptible members. Any rise in toxicity of the films used in the rest of the experiment cannot therefore be ascribed to a fall in resistance of the fly stock during the period of the experiment.

The data for the main part of the experiment are summarised in Table VII.

The data in Table VII support the conclusions which have been drawn from the earlier work, namely, that a given quantity of DDT is best applied to wall-board as a relatively light deposit of a correspondingly concentrated solution, and that with certain combinations of concentration and deposit a sharp increase in toxicity follows the first exposure of the films to flies. On all samples where the toxicity increased after the initial exposure to insects, crystals of DDT could be detected with the naked eye or under the microscope with relatively low magnification (×30). In confirmation of the data of Table II, all deposits from 40 to 320 mg. DDT/sq. ft. laid down from a 5 per cent. solution increased in toxicity ; in confirmation of the data of Table III, the 40 mg. deposit laid down from a 2·5 per cent. solution showed no increase ; neither the 40 nor the 80 mg. deposit laid down from a 1·25 per cent. solution showed an increase. It would seem from these figures that the product of concentration (per cent. w./v. DDT in Pool burning oil) × deposit (mg. DDT/sq. ft.) must exceed a value of about 150, if a marked increase in toxicity to flies due to secondary crystallisation is to be observed under the experimental conditions described. DDT films which did not show the increase in toxicity after the first exposure to flies showed no increase after subsequent exposures.

The persistence of toxicity of the DDT films was also dependent upon the deposit applied and the concentration of the solution used. When no crystals were detected, films which failed to crystallise lost steadily in toxicity and became completely

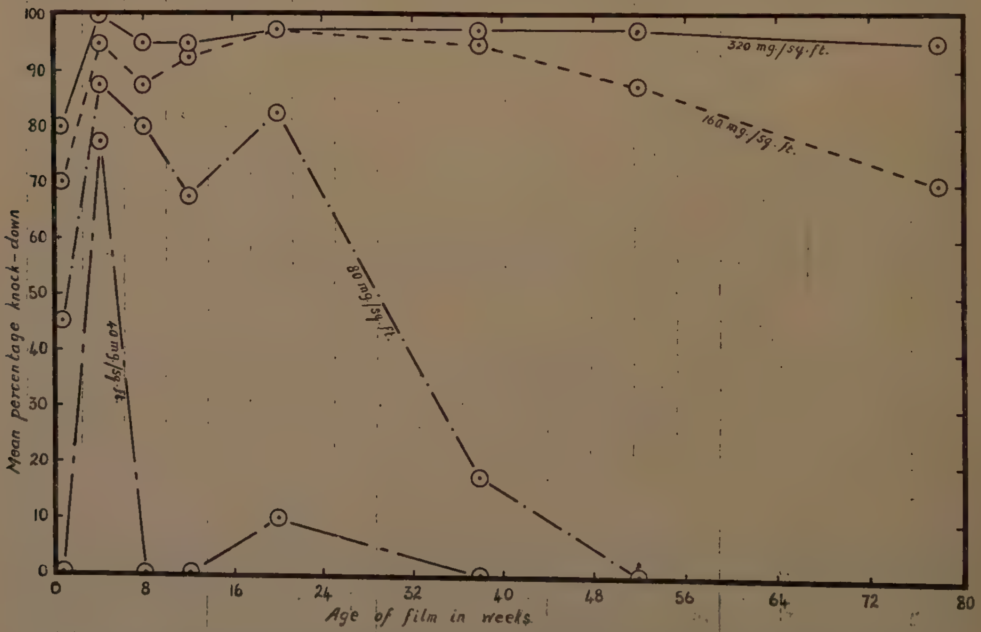

Fig. 1.—Mean percentage knock-down of flies after exposure for 1 hour on wall-board sprayed with different deposits of 5 per cent. DDT in kerosene. Exposures made intermittently during storage of the films at 27·5°C. and 60 per cent. R.H.

ineffective as judged by the percentage knock-down of flies exposed for 4 hours. The persistence of toxicity of the remaining films is best shown graphically, and the data for mean percentage knock-down of flies after exposure for 1 hour are presented

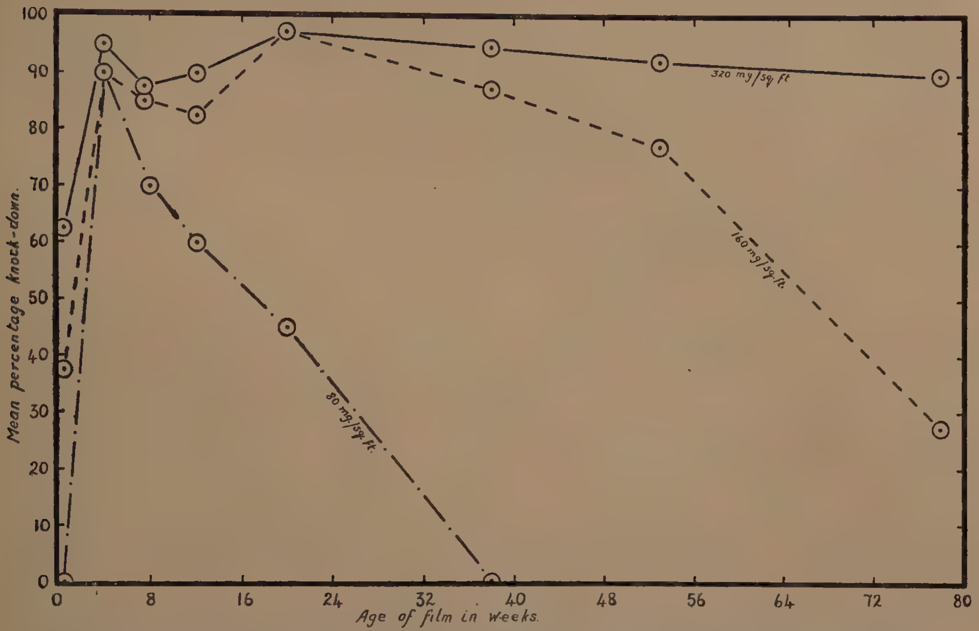

Fig. 2.—Mean percentage knock-down of flies after exposure for 1 hour on wall-board sprayed with different deposits of 2·5 per cent. DDT in kerosene. Exposures made intermittently during storage of the films at 27·5°C. and 60 per cent. R.H.

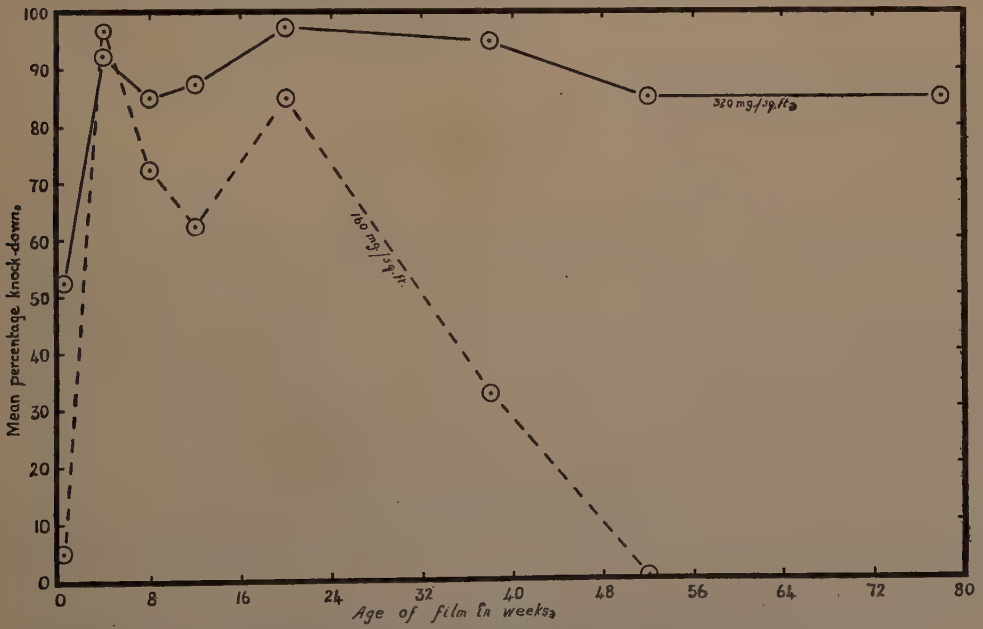

Fig. 3.—Mean percentage knock-down of flies after exposure for 1 hour on wall-board sprayed with different deposits of 1·25 per cent. DDT in kerosene. Exposures made intermittently during storage of the films at 27·5°C. and 60 per cent. R.H.

in figs. 1, 2 and 3: the three sets of data refer respectively to the 5, 2·5 and 1·25 per cent. solutions of DDT from which the films were laid down. The lines joining the series of points are convenient links only and are not intended to show the course of the toxicity curve between the individual observations.

It will be observed in the figures that films which maintained a relatively high toxicity during at least the first 26 weeks of the test all showed a very distinct increase in toxicity between the exposures to flies at 4 days and 4 weeks. Each film then lost in toxicity during the second 4 weeks of storage. Between the eighth and twelfth weeks, the more effective group of films gained a little in toxicity, whereas the less effective group lost a little. After a further 8 weeks of storage, in which there was no intermediate test with flies, both groups showed a definite increase in speed

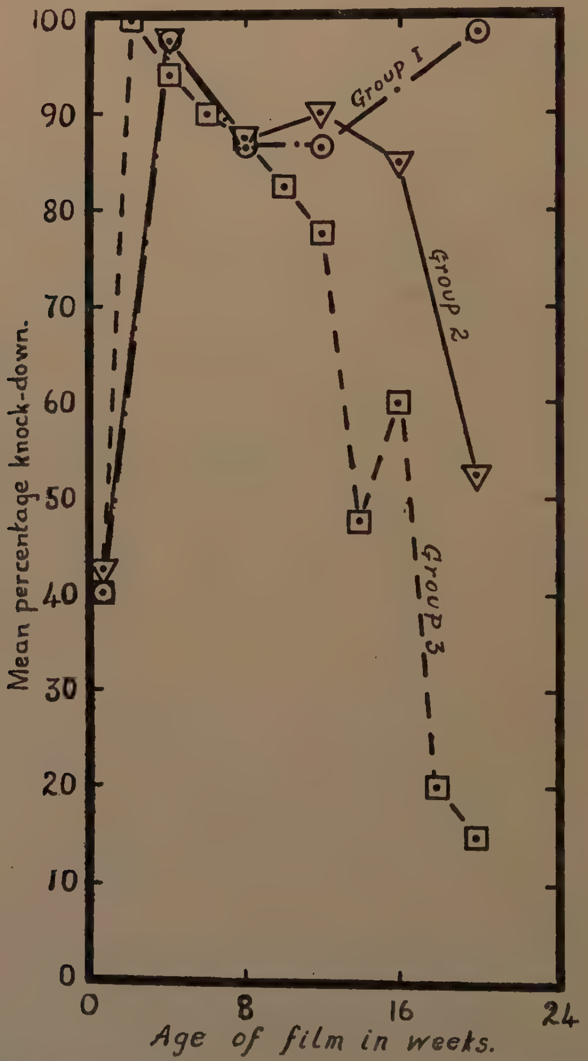

Fig. 4.—Mean percentage knock-down of flies after exposure for 1 hour on wall-board sprayed with 80 mg. DDT/sq. ft. from 5 per cent. solution in kerosene. The graphs show the effect on toxicity of different intervals between exposures to flies.

of action. From the twentieth week onwards effectiveness decreased at different rates according to the combination of concentration and deposit originally applied.

The rise in toxicity between the twelfth and twentieth weeks was considered due to partial or complete regeneration of the crystal carpet. Such powers of regeneration were, however, obviously limited. The occurrence of this phenomenon between the twelfth and twentieth weeks of the test was ascribed to the fact that the samples had a longer interval between exposure to flies at this stage of the experiment than earlier, and another experiment was started to test this assumption.

Three groups, each of four squares of wall-board, were sprayed with 80 mg. DDT/sq. ft. from a 5 per cent. solution. All were exposed to flies on the fourth day after treatment. Thereafter, the samples were exposed to flies as follows :—

Group 1—after 4, 8, 12 and 20 weeks.

Group 2—after 4, 8, 12, 16 and 20 weeks.

Group 3—after 2, 4, 6, 8, 10, 12, 14, 16, 18 and 20 weeks.

The data representing the mean percentage knock-down of flies after exposure for 1 hour on each group of boards are shown in fig. 4 plotted against the age of the film.

The samples of Group 1 were exposed to flies at the same time intervals as were used in the main experiment (see figs. 1–3) and the toxicity of the DDT film rose and fell in approximately the same manner ; there was a definite rise in toxicity in the interval between the exposure at the twelfth and twentieth weeks. The samples of Group 2 had an additional exposure at the sixteenth week. The toxicity of the films remained fairly constant at the eighth, twelfth and sixteenth weeks, then fell sharply at the twentieth week. The Group 3 samples, which were exposed at bi-weekly intervals throughout the course of the experiment, fell at each exposure after the initial increase between the fourth and fourteenth days, except for a rise between the exposures at 16 and 18 weeks, for which no explanation can be advanced. The data in general, however, support the assumption that the rise in toxicity of the films between the twelfth and twentieth weeks in the main experiment was due to a partial regeneration of the carpet of DDT crystals during this extended interval between exposures to flies.

Discussion.

In the period during which this work has been in progress a number of papers have been published reporting that the toxicity of residual deposits of DDT on different surfaces can persist for a year or more. Relatively few papers, however, have dealt with the factors governing the formation of effective films. In the present work the expected importance of two factors, concentration of DDT in the original kerosene solution and deposit of DDT applied to the surface, has been demonstrated, using wall-board as the experimental substrate. The greatest toxicity from a given amount of DDT will result from the application of the insecticide in as concentrated a solution as is consistent with reasonable coverage of the surface, but it is not yet known whether a continuous film is preferable to a discontinuous deposit. The data accumulated from the experiments on wall-board indicate that the DDT should be applied in such a way as to leave as much as possible on the surface of the substrate and this point will presumably be of equal importance in the treatment of other absorbent materials. The application of a solution of DDT in kerosene is probably by no means the best method of achieving this superficial deposition under many practical conditions, but has advantages when an inconspicuous film is wanted or damage would result from the use of water as the carrier.

The marked increase in toxicity of DDT films on wall-board, formed by a combination of concentration and deposit above a certain critical value, following the first exposure to flies has not been reported by other workers using different substrates. It seems clear that this increase is due to crystallisation of DDT from the supersaturated solution left after evaporation of the volatile constituents of the spray. The stimulus for crystallisation of the supersaturated residue may be any form of mechanical agitation, including movement of flies on the treated surface. This phenomenon has not yet been observed on other absorbent materials used in the latter part of our investigation, although it can be demonstrated on glass. It may be that the effect is peculiar to wall-board among absorbent materials but, more probably, the correct combinations of concentrations and deposits of the DDT solutions in relation to the critical value for each material have not been used.

The experiments described here have been carried out under laboratory conditions, probably highly favourable for persistence of toxicity of DDT films, inasmuch as the treated samples have been stored in a relatively dust-free atmosphere and in the absence of sunlight. Equal persistence of toxicity can therefore not be expected under practical conditions, but plenty of evidence has already been obtained to show that DDT-in-kerosene solutions sprayed on to different surfaces in kitchens, factories, stables, etc., are sufficiently persistent to be of the greatest practical importance.

Summary.

Exposure of house-flies to residual films of DDT, deposited on wall-board from kerosene solutions, has shown that the initial toxicity increases (1) as the deposit of DDT per unit area increases, and (2) as the concentration of DDT in the solution increases, although the amount of DDT applied is kept constant. In practice, DDT in kerosene should be applied to absorbent materials at as high a concentration as possible, consistent with reasonable coverage, in order to avoid much of the DDT being carried by the solvent into the substrate, where it is no longer available for contact with the insects.

With wall-board, a critical value for the product of concentration and deposit of DDT has been shown to exist, above which any mechanical stimulus, including movement of flies on the treated surface, will cause the DDT present in supersaturated solution to crystallise, with consequent marked increase in toxicity of the film. Under some circumstances, removal of the crystal carpet is followed by its partial regeneration. If the product of the concentration and deposit falls below the critical value, the residual film is at no time very effective and soon loses its initial toxicity.

The persistence of toxic films of DDT on wall-board has been shown, over an exposure period of 18 months, to be closely bound up with the crystallisation of the deposits and their powers of regeneration during intermittent exposure to flies.

This work has been carried out as part of the programme of research of the Pest Infestation Laboratory, and this account is published by permission of the Department of Scientific and Industrial Research.

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THE USE OF BAMBOO POTS TO INDICATE *AËDES* PREVALENCE.*

By K. S. Hocking, A.R.C.Sc., B.Sc.

22

Introduction.

Although the importance, throughout Africa, of maintaining aerodromes free from *Aëdes* is generally realised, no satisfactory method of assessing *Aëdes* populations on rural airfields, not surrounded by houses, has been described. It was, therefore, thought desirable to publish the results of attempts to assess the efficiency of *Aëdes* controls by means of bamboo pots, even though they may be scanty and unsatisfactory. Despite their incompleteness these results do still suggest that bamboo pots may be used to give a reliable index of *Aëdes* prevalence in rural areas, and to provide also information not given by the House Larval Index in urban areas.

Harris (1942), acting on the experience of Dunn (1927) in West Africa, exposed bamboo pots in batches of 25 for 5 days at a time, as artificial breeding places to indicate *Aëdes* prevalence, in three different areas in the Morogoro district of Tanganyika.

His results were briefly as follows :—

1. In Morogoro Township. The percentage of pots containing larvae after five days' exposure was 80 per cent. in shaded places and 40 per cent. in unshaded places in May/June and fell to 20 per cent. shaded and 12 per cent. unshaded in September. The corresponding township House Larval Index was 2·0 in April and nil in August.
2. In the centre of Morogoro Airfield, 0·7 miles from the edge of the township. Mean percentage of pots with larvae from May to July—10 per cent. *A. aegypti*, L., 17 per cent. *A. simpsoni*, Theo. Mean percentage in township for same period—38 per cent. *A. aegypti*, 33 per cent. *A. simpsoni*.
3. Densely populated rural area on mountainside above Morogoro. Percentage of pots with *A. aegypti* larvae was 0 in May and rose to a peak of 60 per cent. in August.

These results of Harris' seem to indicate that this method may be of value in assessing an *Aëdes* population, particularly in rural areas where a House Larval Index is impossible or meaningless.

During the war *Aëdes* controls were started in several such rural areas in East Africa, particularly to protect aerodromes, and an endeavour was made to assess the efficiency of these controls by the use of bamboo pots.

The main results are from four places :—

Antsirane, the town of Diego Suarez, Madagascar	July 1943–August 1945.
A rural camp at Sakaramy near Diego Suarez	December 1943–October 1944.
Port Reitz Aerodrome, Mombasa, Kenya	January 1945–December 1945.
Dar-es-Salaam Aerodrome, Tanganyika	February 1945–December 1945.

Method.

Bamboo pots as described by Harris, of about 3 in. internal diameter and about 1 ft. in length, were filled with water and exposed in groups of varying numbers (usually ten). Pots smaller than this were found to be unsatisfactory and new pots were unsatisfactory for the first two or three weeks.

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The pots were examined after five days' exposure and any larvae found were recorded. The larvae were sometimes identified and sometimes merely recorded as *Aëdes* sp. or *Culex* sp. The pots were then washed out with hot water, dried and replaced after a two-day interval.

Pots were generally 3 ft. from the ground, but sometimes at ground level and sometimes at 6 ft.

Results.

Antsirane.

Date	No. of Pots exposed	Percentage positive	House Larval Index	Rainfall in inches
1943				
April	40	62	—	0·51
July	301	12	7·2	0·91
August	211	3·8	—	0·12
September	355	2·8	0·9	0·02
October	343	6·4	1·6	0·28
November	282(control	11·0	2·4	0·28
December	360 began)	11·4	2·5	2·28
1944				
January	292	18·2	6·9	9·6
February	144	24·3	6·2	4·33
March	280	26·4	3·9	1·14
April	200	30·0	1·9	2·04
May	134	18·7	1·2	0·11
June	160	21·9	0·8	0·15
July	80	38·0	0·9	0·43
August	80	12·5	0·6	0·23
September	80	6·0	0·8	0·11
October	80	1·3	0·3	Nil
November	80*	5·0	0·3	0·19
December	68*	8·8	0·4	6·25
1945				
January	48*	29	1·46	9·3
February...	48*	29	1·81	6·6
March	48*	25	1·15	2·5
April	80	11	1·18	1·9
May	80	2·5	0·74	1·7
June	100	0	0·26	0·3
July	80	0	0·17	Nil
August	80	0	0	1·7

*Pots exposed at Lazaret Point 1 mile outside town.

Sakaramy.

Date	No. of Pots exposed	Percentage positive
1943		
December	405	4·2
1944		
January	320	63·4
February	316	75·6
March	391	49·9
April	286	24·1
May	239	11·3
June	193	3·6
July	206	2·9
August	188	0·5
September...	188	0·5
October	129	0

Sakaramy is about 15 miles inland from Antsirane. The rainfall there is rather heavier and the season lasts rather longer.

Port Reitz Aerodrome, Mombasa.

Date	No. of Pots exposed	Percentage positive	House Larval Index	Rainfall in inches
1945				
January	84	1·2	0·57	0·47
February	84	4·8	0·4	trace
March	105	4·8	0·57	1·3
April	Pot size increased from 1 to 3 ins.			
			0·48	5·9
May	180	19·4	0·9	19·25
June	296	36·8	0·88	1·85
July	230	41·7	0·51	2·7
August	240	45·0	0·26	1·21
September	300	20·7	0·17	2·52
October	216	11·1	0·29	0·74
November	109	10·1	0·33	8·16
December	300	18·7	0·38	0·34

Dar-es-Salaam Aerodrome.

Date	No. of Pots exposed	Percentage positive	House Larval Index	Rainfall in inches
1945				
February...	Pots first exposed mid-February		0·31	2·18
March	450	3·8	0·45	5·95
April	360	7·2	1·01	7·29
May	360	3·1	0·31	8·63
June	450	0·9	0·07	0·40
July	360	0	0·06	0·42
August	360	0	0·03	0·75
September	450	0	0·05	1·75
October	360	0	0·05	4·04
November	360	0	0·12	2·4
December	450	0	0·09	1·4

Discussion.

The theoretical meaning of the indices.

The *Aëdes* House Larval Index is the percentage of houses containing *Aëdes* larvae. If it is accurately recorded, it is, in effect, a direct measure of the number of egg-laying females about during each cycle of inspection, until the number of possible breeding places becomes so low that some females cannot find a place in which to lay their eggs.

The Pot Index is the percentage of the bamboo pots exposed that contain *Aëdes* larvae at the end of the period of exposure. With a fixed number of egg-laying females it will vary according to the number of breeding places available to the females. In other words, a constant number of pots is a varying proportion of the total breeding places available to the egg-laying female mosquitos. For example, assuming the bamboo pots and all other breeding places are equally attractive to the mosquitos, 100 females laying eggs in an area containing 100 pots and 900 other breeding places will, on an average lay in 10 of the pots giving a Pot Index of 10 per cent.; but, if the area contains only 100 other breeding places they will lay in 50 of the pots and produce a Pot Index of 50 per cent.

Application to the Results.

Using these interpretations of the Indices, the results may be used to build up a picture of the varying *Aëdes* populations and consequently of the efficiency of the various controls. At the same time, by the reasonableness or otherwise of the picture built up, an indication may be obtained of the value of this method of assessment.

Antsirane.

From Antsirane there is a long series of results, but the number of pots was small and concentrated in one area, and for five months (November 1944–March 1945) they were exposed one mile outside the town.

Immediately following the break of the rains in December 1943, the Antsirane House Larval Index rose to a peak of 6·9 in January 1944, and immediately thereafter fell steadily to 0·8 in June 1944. The Pot Index also rose following the rain, owing to the increase in adult population, but during the period of falling adult population from January to April, as indicated by the falling House Larval Index, the Pot Index continued to rise (to a maximum of 30 per cent. in April) because of the rapidly decreasing number of natural breeding places. With the almost complete dying away of the adults after June 1944, the Pot Index also fell to a minimum in October.

The *Aëdes* control that had been carried out in Antsirane from October 1943 onwards consisted essentially of removing or destroying potential breeding places; and the combination of Pot Index and House Larval Index can be used to estimate the numbers of such potential breeding places. As the bamboo pots were exposed in only one place in the town it is unsafe to use this method of estimation when the number of larval collections (and hence the number of egg-laying females) falls below say 10 per week, which limits the period at Antsirane to July 1943–July 1944. (During the rains 1944–45, pots were not exposed in the town itself.) Assuming the pots are a fair sample of the breeding places available, *i.e.* that the percentage of pots with larvae is the same as the percentage of all other breeding places with larvae, the total collections of larvae determined from the House Larval Index can be used with the Pot Index to give the total number of breeding places available. For example, a House Larval Index of 1 per cent. means that one house in 100 contains larvae and that in 4,000 houses there would normally be 40 collections of larvae. If at the same time the Pot Index was 10 per cent., those 40 collections of larvae may be assumed to be 10 per cent. of the breeding places available to the mosquitos in the area covered by the House Larval Index. These breeding places would therefore number 400.

On this basis the total numbers of breeding places available in Antsirane from July 1943 to July 1944 were as follows :—

Date	House Larval Index	Larval collections in 4,000 houses	Pot Index	Total breeding places available	Rainfall
1943	Per cent.		Per cent.		
July	7·2	288	12	2,400	0·91
August			3·8		0·12
September	0·9	36	2·8	1,300	0·02
October	1·6	64	6·4	1,000	0·3
November	2·4	96	11·0	900	0·3
December	2·5	100	11·4	900	2·3
1944					
January	6·9	276	18·2	1,500	9·6
February	6·2	248	24·3	1,000	4·3
March	3·9	156	26·4	600	1·1
April	1·9	76	30·0	250	2·0
May	1·2	48	18·7	260	0·1
June	0·8	32	21·9	150	0·2
July	0·9	36	38·0	100	0·4

The numbers are obviously very approximate but do suggest that with larger numbers of pots distributed throughout the town the method could be used to give information that the House Larval Index alone could not give. For example, in September 1943, before control measures had started, the House Larval Index was the same as in July 1944, after nearly a year of control measures; but the numbers of breeding places, as given in the table, are very different (1,300 and 100).

In July 1944, the number of pots exposed was reduced from 160 to 80. In the 80 pots and the 100 other available breeding places (in 4,000 houses) there were 66 collections of larvae. If the number of pots had remained 160 and these 66 collections of larvae had been distributed among the 160 pots and the 100 other potential breeding places, the Pot Index would have been 26 per cent. and the House Larval Index 0·6 per cent. instead of 0·9 per cent. When the number of available breeding places is small the number of pots exposed can appreciably affect the House Larval Index.

Sakaramy.

In the rural area of Sakaramy, where no House Larval Index was available, the sequence of events can be followed by analogy with the Antsirane results.

In mid-December 1943, it can be assumed that the breeding places had been increased by the rain but that the adult mosquito population had not yet increased, and the Pot Index was low (4·2 per cent.). By mid-January 1944, the adult population had increased greatly without any appreciable further increase in the number of breeding places, and the Pot Index rose sharply (to 63 per cent.). The adult population was maintained, or even increased, until mid-February, by which time the number of breeding places had fallen, and the Pot Index rose again (to 75 per cent.). The ratio of breeding places to adult mosquitos apparently fell more slowly than in Antsirane (possibly because of the greater amount of bush at Sakaramy and the control measures in Antsirane), and the Pot Index fell from March onwards to nil in October.

Port Reitz, Mombasa.

Large numbers of pots were distributed throughout this area, but for a short time only—in effect, from the height of the long rains in 1945 through the dry season to the short rains at the end of 1945—but they give a similar picture of events to that at Diego Suarez.

The Pot Index was 19 per cent. in May with a large adult mosquito population following rain in April and May, but as the outside breeding places dried up, the Index rose for three months to a peak of 45 per cent. in August and then (the numbers of adults having become very low) it fell to 11·1 per cent. in October. In November the short rains started and with the increase in the number of breeding places the index fell to 10 per cent. December was dry again, and the number of adults having increased, the index rose to 19 per cent.

The House Larval Indices from the adjoining mainland area of Mombasa Township, in conjunction with the Pot Indices, can be used to estimate the numbers of potential breeding places in the same way as before.

The " Mombasa Mainland " is a rural area and the House Larval Index is therefore based on only a small proportion of the actual collections of larvae in the area. Also, it is not strictly comparable with the Port Reitz Aerodrome area, although it is sufficiently so for it to be reasonable to assume that a House Larval Index from the Port Reitz area would be very similar. The estimated numbers of potential breeding places are, therefore, even more approximate than those calculated for Antsirane and can give no more than an indication of the trend of changes.

Date	Mainland House Index	Larval collections from 10,000 houses	Pot Index	Potential breeding places	Rainfall inches
1945	Per cent.		Per cent.		
April	0·48	48	—	—	5·90
May	0·90	90	19·4	470	19·25
June	0·88	88	36·8	240	1·85
July	0·51	51	41·7	120	2·70
August	0·26	26	45·0	60	1·21
September	0·17	17	20·7	80	2·52
October...	0·29	29	11·1	261	0·74
November	0·33	33	10·1	327	8·16
December	0·38	38	18·7	203	0·34
1946					
January			25·6		0·02

Dar-es-Salaam.

On Dar-es-Salaam Aerodrome large numbers of pots were exposed throughout the area from early in the long rains through the dry season and the short rains at the end of 1945, but the *Aëdes* population was apparently so low (possibly owing to the open nature of the bush cover) that the Pot Index never rose above 7·2 per cent. The bamboo pots used were not more than two inches in diameter and the index would undoubtedly have been higher if the pots had been the usual three inches (compare the low Pot Index in Mombasa before April, 1945).

Practical Value of the Indices.

As explained above the House Larval Index is a measure of the adult population only if it is a completely accurate record. In practice its degree of accuracy is rarely very high and may be low. This may be because of the inaccessibility of some *Aëdes* breeding places, the difficulty of searching some places thoroughly, or the lack of diligence of some of the searchers. The practical discrepancy between the House Larval Index and the number of *Aëdes* adults is illustrated by the findings of McCarthy and Wilson (to be published shortly) that dengue transmission continued in Antsirane until the House Larval Index fell below 2 per cent., although in Mogadishu it ceased when the House Larval Index was 10 per cent.

The Pot Index is not subject to these practical limitations and, if the pots are sufficiently numerous and well distributed throughout the area, very little supervision is necessary to ensure an accurate record. The Pot Index depends on the proportion of adults to possible breeding places and in a naturally fluctuating *Aëdes* population it may be difficult to interpret ; but, in a well controlled area, where the number of possible breeding places is very low and more or less constant, the Pot Index gives a direct measure of the adult population and a sudden emergence or influx of adults would be followed by a sudden rise in the Pot Index.

The main practical value of the Pot Index seems to be therefore :—

i. To be used with the House Larval Index to give a comprehensive assessment of *Aëdes* fluctuations, and *Aëdes* control measures.

ii. To give a sensitive index of adult numbers in a well controlled area.

iii. To give information about *Aëdes* prevalence in a rural area where a House Larval Index is meaningless or impossible.

Summary.

1. The method used is founded on those of Dunn (1927) and Harris (1942).

2. Results are given for 1943, 1944 and 1945 from areas in Kenya, Tanganyika and Madagascar.

3. An attempt is made to interpret these results in the light of the theoretical meanings of the Pot Index and the House Larval Index. This demonstrates that these two indices together can be used to assess the fluctuations in an *Aëdes* population and hence the efficiency of control measures.

4. Finally, the practical value of the Pot Index is discussed.

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PRELIMINARY NOTES ON THE LOSS OF DDT AND GAMMEXANE BY ABSORPTION.

By F. Barlow and A. B. Hadaway.
Colonial Insecticide Research, Uganda.

Q.C.

Introduction.

During the course of investigations into the use of the insecticides DDT and Gammexane, it has become increasingly clear that there is considerable loss through absorption into the treated surface. This loss of contact-available insecticide is most marked when simple oil solutions are used. Dosages of 100 to 200 mgms. per sq. ft., which on a non-absorbent surface (*e.g.* glass) are 100 per cent. toxic to flies exposed for very short periods, show little toxicity when applied to absorbent surfaces (*e.g.* mud).

The tests summarised here have been carried out in Uganda during field and laboratory investigations with tsetse (*Glossina palpalis*, R.-D.) on the shores and islands of Lake Victoria, and with mosquitos in native huts.

It must be emphasised that they are preliminary results that require confirmation but it is thought that they may suffice to indicate the problem and the need for much serious study.

Tests against Tsetses.

Application to Vegetation.

Two types of plants were selected for sampling, *Alchornea* and *Triumfetta*, both of which are common in the lake shore bush inhabited by tsetse. The leaves of *Alchornea cordata* (Euphorbiaceae) are smooth and hard, whilst those of *Triumfetta macrophylla* (Tiliaceae) are soft and very hairy.

In nearly all field tests routine chemical and biological tests have been made on these leaves taken at random from treated bush, which, of course, is totally exposed to climatic conditions. In addition, a small series of estimations of surface deposits on leaves of Avocado pear, which are smooth and hard, and of *Hibiscus* which are fairly soft and hairy, has been made.

For the chemical estimations, leaves were first stripped with benzene to remove surface deposits. They were then dried, powdered and extracted in a Soxhlet apparatus for the absorbed insecticide. In preliminary work leaves of approximately 30 sq. in. were washed with two lots of 20 cc. of benzene followed by a further 20 cc. and these two extracts analysed separately. It was found that the first extract with two portions of benzene was adequate for all the insecticide preparations used, and also that the relative values found inside and outside the leaf did not depend upon the extent of washing. All estimations are based upon the amounts of hydrolysable chlorine present.

Biological tests have consisted of simple contact exposures of laboratory bred *G. palpalis* to the treated leaf surfaces. Usually batches of 20 flies were exposed individually for 20 secs.

(i) Known dosages of DDT (pure pp′) were applied accurately by means of a pipette as a solution in benzene to leaves of Avocado pear and *Hibiscus*, which were exposed outside to ordinary climatic conditions. Chemical determinations of deposits were made on sample leaves from time to time; results are shown in Table I. There appears to have been little loss by absorption into the leaves; 80 to 90 per cent. of DDT was recovered from the surface of the leaves four to six days after application. This is probably due to the high volatility of the solvent, and the consequent immediate deposition of solid DDT on the leaf surface.

Deposits were lost much more rapidly from the soft *Hibiscus* than from the hard-surfaced Avocado pear leaves.

TABLE I.

Longevity of DDT deposits on leaves of Hibiscus *and Avocado pear exposed outside to ordinary climatic conditions.*

Dosage applied	Recoveries of DDT in mgms./sq. ft. on various days after the application of insecticide						
	4	5	6	14	15	42	72
Hibiscus—							
50 mgms. per sq. ft.	—	—	42	—	24	—	—
100 " "	62	—	55	—	58	—	—
200 " "	—	145	—	—	107	—	—
Avocado pear—							
50 mgms. per sq. ft.	—	—	44	40	—	37	36
100 " "	—	—	94	90	—	80	—
200 " "	—	—	—	191	—	119	—

(ii) During a field trial on Mbirubuziba Island, treated leaves were taken at random over a period. After determining their toxicity to *G. palpalis*, chemical estimations of deposits both inside and outside the leaves were made. Results in Table II show that about 20 per cent. of the insecticide recovered was inside the leaf.

There is no close correlation apparent between estimated surface deposits and toxicities to flies. In fact, results, as seen in the tables, are not infrequently bewildering. Two factors are probably mainly responsible, the individual reactions of flies and the method of testing samples for toxicities and extracting for estimations of deposits. Batches of flies are usually exposed to two or three leaves taken at random from a sample of leaves sprayed in the field. Deposits vary not only from leaf to leaf but also from one part of a leaf to another. Each fly of the batch is placed on a contact site at random. Some may by chance stand on an ample deposit ; others may stand on a scanty one. The chemical estimation is based on extraction of deposits from the total surfaces of the two or three leaves. It is therefore an average of a very large number of possible contact-sites, a few only of which were used for exposures of flies.

Close correlation of chemical and biological results may be obtained when it is possible to use much bigger batches of flies in laboratory tests.

TABLE II.

Toxicity to G. palpalis *of leaves treated with* 5 *per cent. DDT* (83 *per cent. pp'*) *solution in* 50 *per cent. kerosene and* 50 *per cent. cotton seed oil on Mbirubuziba Island, and recovery of DDT from the same leaves.*

Leaves used	No. of days after spraying	Recovery of DDT in mgms/sq.ft.		Percentage kill of *G. palpalis* 24 hours after exposure for 10 seconds
		Outside of leaves by washing	Inside of leaves by extraction	
Alchornea	1	114	Not determined	67
"	6	130	31	—
"	6	96	22	—
"	10	58	8	48
"	16	113	29	5
Triumfetta ...	10	77	8	13
" ...	16	34	4	15

(iii) In a later trial on Mbirubuziba Island a patch of vegetation was sprayed to give as uniform a deposit of insecticide as possible. Sample leaves were taken at intervals for chemical and biological tests, results of which are shown in Table III. Climatic conditions on the island during this trial are given in Table IV.

The initial loss by absorption into the inside of the leaves was rapid and great, reaching in one case over 50 per cent. of the total insecticide recovered. This was followed by a gradual loss of insecticide both from the outside and the inside of the leaves. The former loss is probably due to various factors such as rain and brushing off by mechanical means. The question arises whether the plant plays an active part in any loss by absorption after the initial penetration, and in the subsequent disappearance of insecticide from the inside of the leaves.

The soft leaves of *Triumfetta* retained the surface deposit longer than did the hard leaves of *Alchornea*. This deposit, however, was much less toxic to flies than was expected, and it is wondered if the hairiness of the leaves prevents, to some extent, contact between the flies and the deposit on the leaf surface.

TABLE III.

Toxicity to G. palpalis *of leaves treated with* 5 *per cent. DDT solution in* 50 *per cent. power kerosene and* 50 *per cent. cotton seed oil on Mbirubuziba Island and recovery of DDT from the same leaves.*

Leaves used	No. of days after spraying	Recovery of DDT in mgms./sq.ft.		Percentage kill of *G. palpalis* 24 hours after exposure of 20 seconds
		Outside of leaves by washing	Inside of leaves by extraction	
Alchornea	1	100	59	100
	3	66	50	68
	5	72	29	35
	8	80	35	25
	13	48	28	25
	21	28	12	10
	25	22	8	10
	28	14	7	25
	35	13	9	10
Triumfetta ...	1	61	65	45
	5	60	26	40
	8	60	11	15
	13	60	7	10
	21	92	15	20
	25	23	2	30
	28	47	7	15
	35	29	4	5

TABLE IV.

Climatic conditions on Mbirubuziba Island during the trial recorded in Table III.

	Mean daily temp. °F.		R.H.			Rainfall in inches
	Max.	Min.	6 a.m.	12 noon	6 p.m.	
1st week	76·0	64·0	93·0	84·7	77·6	0·12
2nd „	79·0	63·0	92·0	72·9	80·4	0
3rd „	75·5	64·2	93·9	80·7	78·4	0·08
4th „	77·4	63·8	94·8	76·3	78·7	0·14
5th „	74·5	63·2	94·9	79·4	77·9	0·6

(iv) Patches of vegetation were sprayed as evenly as possible with various preparations of DDT and Gammexane. It was not possible to conduct chemical and biological tests on identical leaves ; the results given in Tables V and VI are not, therefore, strictly comparable.

Again there was as much as 50 per cent. loss of both DDT and Gammexane through absorption into the leaves when simple solutions were applied. A much lower proportion of DDT was recovered from the inside of the leaves treated with ATSO emulsion, and only a very small fraction of the Gammexane applied as a water suspension.

Rapid disappearance of Gammexane from both the outside and inside of the leaves occurred in all cases.

Climatic conditions during the test are shown in Table VII.

TABLE V.

Chemical estimations of insecticide deposit on and in leaves after spraying with various preparations.

Formulation	Leaves used	Mgms./sq. ft. recovered from leaves collected at various times after treatment			
		Few hours	1 week	2 weeks	3 weeks
5 per cent. DDT in power kerosene	*Alchornea* ...	85	Leaves "burned" and fell from plant		
		59			
	Triumfetta ...	98			
		97			
5 per cent. DDT in 50 per cent. power kerosene and 50 per cent. cotton seed oil	*Alchornea* ...	39	67	79	54
		40	43	31	30
	Triumfetta ...	108	57	81	93
		41	32	10	23
5 per cent. DDT emulsion (ATSO)	*Alchornea* ...	41	88	34	38
		18	5	7	26
	Triumfetta ...	127	74	54	44
		21	11	7	5
5 per cent. D.929 in power kerosene, containing 0·5 per cent. gamma	*Alchornea* ...	28	Leaves "burned" and fell from plant		
		9			
	Triumfetta ...	137			
		60			
5 per cent. D.929 in 50 per cent. power kerosene and 50 per cent. cotton seed oil	*Alchornea* ...	59	10	11	3
		54	4	5	2
	Triumfetta ...	67	36	5	13
		18	5	1	2
Gammexane emulsion MG.035 containing 1 per cent. gamma	*Alchornea* ...	20	4	3	3
		8	4	2	1
	Triumfetta ...	12	5	5	4
		13	3	2	1
Water suspension of Gammexane dispersible powder P.530, containing 1 per cent. gamma	*Alchornea* ...	356	166	54	14
		13	5	3	1
	Triumfetta ...	358	310	137	137
		13	9	5	7

The upper figure is the number of mgms. of Gammexane (36 per cent. hydrolysable chlorine) or DDT per sq. ft. obtained by washing the outside of the leaves with benzene ; the lower is the quantity obtained by drying and powdering the washed leaves and then extracting in a Soxhlet.

TABLE VI.

Percentage kill of G. palpalis *exposed for 20 seconds to leaves collected at varying periods after application of insecticide.*

Formulation ...	Leaves used	Age of deposit on freshly collected leaves			
		Few hours	1 week	2 weeks	3 weeks
5 per cent. DDT in 50 per cent. kerosene and 50 per cent. cotton seed oil	*Alchornea* ... *Triumfetta* ...	100 95	10 10	40 15	0 0
5 per cent. DDT emulsion (ATSO)	*Alchornea* ... *Triumfetta* ...	65 60	60 35	55 40	70 0
5 per cent. D.929 in 50 per cent. kerosene and 50 per cent. cotton seed oil	*Alchornea* ... *Triumfetta* ...	100 100	10 30	15 40	0 0
Gammexane emulsion MG.035	*Alchornea* ... *Triumfetta* ...	95 85	25 15	0 0	0 5
Water suspension from Gammexane dispersible powder P.530	*Alchornea* ... *Triumfetta* ...	90 70	90 30	30 0	0 5

These leaves are not identical with those used for chemical estimations given in Table V.

TABLE VII.

Climatic conditions during trial recorded in Tables V and VI.

	Mean daily temp. °F.		Mean R.H.		Rainfall in inches
	Maximum	Minimum	8.30 a.m.	2.30 p.m.	
1st week	83·3	64·4	83·0	57·1	0
2nd ,, ...	82·4	64·0	76·7	57·3	0·03
3rd ,, ...	84·4	64·1	71·1	50·0	0

Tests against Mosquitos.

Application to Mud and Thatch.

The African hut usually has mud walls and a roof of grass thatch. Each of these surfaces presents an interesting problem.

(i) In a preliminary laboratory test, surfaces were treated with a 5 per cent. DDT (83 per cent. pp′) solution in 50 per cent. power kerosene and 50 per cent. cotton seed oil. On application to mud there was rapid absorption, the surface appearing quite dry immediately. Chemical estimation of deposits are shown in Table VIII. The DDT was extracted from the removed layer of mud by shaking with benzene and filtering through cotton wool. Extraction of the residue in a Soxhlet did not yield a further quantity of insecticide.

TABLE VIII.

Days after application	Material	Mgms. DDT per sq. ft.
2	Glass	183
5	Thatch—upper ½ inch 2nd layer of ½ inch	94 20 } 114
6	Mud—upper 1/20th inch 2nd ,, ,, 3rd ,, ,, 4th ,, ,,	92 41 17 } 156 6

G. palpalis, laboratory bred, were exposed individually in tubes for 20 secs. to similarly treated surfaces at various intervals. A summary of percentage kills after 24 hours is given in Table IX.

TABLE IX.

Material	Age of deposit in days				
	1	7	14	23	39
Mud	15	0	0	5	0
Thatch	100	35	5	5	5
Glass	100	100	90	95	85
Controls	0	5	0	0	0

It will be seen that about 40 per cent. of the application to mud was found relatively deeply absorbed. Considerably more than this was lost as contact-available insecticide. This is confirmed by the kills in Table IX.

The loss of contact-available insecticide on thatch may be due not only to obvious penetration of the spray through the interstices to a considerable depth, but also to absorption by the dried grass stems themselves. This has not been investigated.

(ii) Mud blocks were sprayed with emulsions and dispersible powders of DDT and Gammexane. Preparations used were :—

(*a*) Gammexane dispersible powder P.530—water suspension containing 1 per cent. gamma isomer.

(*b*) Gammexane emulsion MG.035 containing 1 per cent. gamma isomer.

(*c*) DDT dispersible powder Guesarol—water suspension containing 1·25 per cent. DDT.

(*d*) DDT emulsion ATSO containing 5 per cent. DDT.

The blocks treated with emulsion dried very quickly showing little evidence of a deposit ; those treated with the suspensions of dispersible powders in water showed a good surface deposit of white particles.

Specimens of *G. palpalis* (laboratory bred) were then exposed to some of these treated mud surfaces at varying periods after the treatment. Results are given in Table XI.

Other mud blocks were used for chemical estimations of deposits. The degree of penetration was found by scraping away successive layers of mud from the surface, extracting with benzene and determining the insecticide in the solution. The quantities recovered are plotted against the weights of the samples of mud taken at each successive " scraping." Results are shown in Table X.

TABLE X.

Chemical estimation of deposits in mgms. of Gammexane or DDT per sq. ft. 28 days after application.

	Mud block treated with		
	P.530	Guesarol	ATSO
Top scraping	38	30	74
2nd "	12	9	56
3rd "	7	2	38
4th "	—	0	21
Total deposits ...	57	41	189

TABLE XI.

Summary of kills of G. palpalis *exposed for* 20 *seconds on mud,* 24 *hours after contact.*

Insecticide	Number of days after application of Insecticide					
	3	17	24	31	38	45
P.530	100	76·7	10	10	0	0
MG.035	5	6·7	0	10	0	0
Guesarol	10	26·7	20	13·3	3·3	6·7
ATSO	0	3·3	0	3·3	0	0
Controls	0	0	0	0	0	0

(iii) In order to compare the toxicities to *G. palpalis* of dispersible powders and simple oil solutions, mud blocks, thatch and glass were sprayed with :—

(*a*) Gammexane dispersible powder P.530—suspension containing 1 per cent. gamma isomer.
(*b*) DDT dispersible powder Guesarol suspension containing 1·25 per cent. DDT.
(*c*) 10 per cent. DDT (83 per cent. pp′) solution in powder kerosene.
(*d*) 5 per cent. DDT (83 per cent. pp′) solution in powder kerosene.

No chemical determinations of deposits were made on these treated surfaces, but deposits were estimated on pilot glass plates, with the following results :—

(*a*) P.530	165	mgms.	Gammexane	per sq. ft.
(*b*) Guesarol	36	,,	DDT	,, ,, ,,
(*c*) 10 per cent. DDT ...	227	,,	DDT	,, ,, ,,
(*d*) 5 per cent. DDT ...	91	,,	DDT	,, ,, ,,

G. palpalis, laboratory bred, were exposed to the treated surfaces individually for 20 seconds at varying periods after the application of the insecticide. Results are given in Table XII. The poor kills of the solutions on mud are undoubtedly due to absorption. The relatively good kills on thatch would suggest that perhaps on this particular type of grass there was little absorption into the grass stems themselves.

TABLE XII.

Percentage kills of G. palpalis 24 *hours after contact.*

Surface	Insecticide	Number of days after application of insecticide							
		1	3	7	9	14	16	21	28
Glass ...	P.530	100					0	0	
	Guesarol	23·3					6·7		26·7
	10 per cent. solution ...	96·7					83·3		93·3
	5 ,, ,, ...	66·7					70		93·3
Mud ...	P.530		93·3		70		16·7	0	
	Guesarol		33·3		36·7		20		16·7
	10 per cent. solution ...		16·7		3·3		0		
	5 ,, ,, ...		10		0		0		
Thatch ...	P.530	80		35		0		0	
	Guesarol	10		0		10		15	
	10 per cent. solution ...	85		80		80		85	
	5 ,, ,, ...	65		100		95		95	
Controls ...		0	0	0	0	0	0	0	3·3

(iv) In a large field trial against mosquitos involving 2,300 huts, mud blocks were placed in sample native huts each of which was sprayed with a different preparation. At various intervals, one block from each hut was brought into the laboratory for exposures of mosquitos and for subsequent chemical determination of the insecticidal deposit. A thin outer layer was scraped from each block, and chemical determinations of the deposit in this layer, and then in the whole block, were made. The weight of the outer layer gives a rough idea of the depth on these blocks; 1 grm. equals approximately ½ mm. over the surface area of 9 sq. in. Determinations so far completed are given in Table XIII.

TABLE XIII.

Preparation	Age of deposit	Weight of outer layer in grms.	Mgms./sq. ft. in outer layer	Total recovery insecticide mgm./sq. ft.	Percentage of total recovery in outer layer
Gammexane dispersible powder P.530	1 day	1·3	5	38	13
	2 weeks	0·9	3	—	—
	4 ,,	2·5	47	185	27
	7 ,,	2·3	34	113	30
Gammexane D.939 solution in dieseline	1 day	1·2	8	252	3
	2 weeks	1·2	4	114	3
	4 ,,	2·0	3	268	4
	7 ,,	2·0	8	196	4
5 per cent. DDT solution in dieseline	1 day	1·0	10	185	5
	2 weeks	1·3	16	196	8
	4 ,,	1·5	21	220	10
	7 ,,	2·3	42	482	9
5 per cent. DDT solution in 50 per cent. power kerosene and 50 per cent. cotton seed oil	1 day	1·3	40	239	17
	2 weeks	1·7	52	374	14
	4 ,,	1·5	44	343	13
DDT dispersible powder Guesarol	1 day	2·5	78	112	69
	2 weeks	3·2	89	273	33
	4 ,,	2·5	94	181	52
	7 ,,	2·8	48	134	36

Guesarol dispersible powder was absorbed to a much less degree than other preparations used. The dieseline solutions were almost entirely absorbed. It seems that the cotton seed oil added to kerosene checks absorption to a small extent.

It is obvious from these tests that there is considerable loss of insecticide when applied to mud as a simple solution or as an emulsion. The dispersible powder Guesarol contains only 5 per cent. DDT, so that suspension in water cannot contain more than 1 or 2 per cent. DDT. Its effectiveness compared with that of emulsions and solutions containing 5 per cent. DDT or Gammexane cannot therefore be strictly determined. Results are, however, promising. The Gammexane dispersible powder was highly effective for two or three weeks, but then lost its toxicity.

Application to Limewash.

The limewashed interiors of many European houses in the tropics also present a problem.

(i) Three blocks of limewashed cement and plaster were cut from old buildings. One block had been limewashed 6 months before, one 9 months, and one 18 months before. They were sprayed with 5 per cent. DDT in power kerosene. *G. palpalis* was exposed to them 48 hours later for 20 seconds, with the following results.

Age of block	Percentage kill in 24 hours	DDT deposit recovered
6 months ...	5	162 mgms. per sq. ft.
9 ,, ...	0	192 ,, ,,
18 ,, ...	0	114 ,, ,,

(ii) Five per cent. DDT in power kerosene was sprayed on a limewashed glass plate 3 hours after application of lime. Kills of *G. palpalis* exposed for 20 seconds were as follows :—

Age of deposit	Lime and DDT (236 mgms./sq. ft.)	Lime only	DDT only (173 mgms./sq. ft.)
24 hours	10 per cent.	0	90 per cent.
48 ,, ...	7 ,,	0	96 ,,

(iii) Limewashed plates of varying ages from 3 hours to 8 weeks were treated with 5 per cent. DDT in power kerosene. Tests were conducted at 24 hours, 7 days and 9 days after the DDT was applied. Kills were never more than 50 per cent., usually less than 25 per cent. Controls, *i.e.* flies exposed to DDT only, gave 100 per cent. kills (except in one instance 93·3 per cent.). DDT deposits recovered from the limewashed plates varied from 64 to 312 mgms. per sq. ft.

(iv) Extraction was made from glass plates which had been limewashed and treated with DDT and which subsequently showed little or no toxicity to flies exposed for 20 seconds. A crystalline compound was obtained having a melting point of 107°C. Twenty mgms. were dissolved in ether and applied to a glass plate to give a dosage of 80 mgms./sq. ft. *G. palpalis* were exposed to this for 20 seconds, and a kill of 83·3 per cent. in 24 hours was obtained. It seemed clear, therefore, that no change was occurring in the DDT applied to limewash, but that loss of toxicity was probably due to masking by absorption.

(v) Longer exposures (2 and 5 minutes) of *palpalis* to treated limewash gave 60 per cent. and 80 per cent. kills. Two-hundred and forty mgms. of DDT per sq. ft. were recovered from this plate. A pilot glass plate treated at the same time with DDT only (152 mgms. per sq. ft.) gave 100 per cent. kills after 20 seconds contact.

Aëdes aegypti, L., were exposed to the same plate. Results are given in Table XIV.

TABLE XIV.

Age of deposit	Exposure	Percentage kill 24 hours after contact	
		To limewash and DDT (240 mgms. per sq. ft.)	To DDT only (152 mgms. per sq. ft.)
9 days	5 mins.	20	100
	10 ,,	40	—
14 days	1 min.	—	100
	10 mins.	33	—
	30 ,,	60	—

(vi) Dispersible powders of DDT and Gammexane sprayed on limewash appear to be more promising than solutions, but sufficient data are not yet available to draw definite conclusions.

(vii) In previous tests the limewash was applied to the glass plates as a thin layer, much thinner than applied in practice to the plaster or cement walls of buildings. A layer approaching this in thickness was applied to a glass plate which was then treated heavily with a 5 per cent. solution of DDT in kerosene. A week later this plate gave a kill of 21 per cent. to *G. palpalis* exposed for 20 secs. Chemical determination showed a deposit of 909 mgms. of DDT per sq. ft.

It seems from these small investigations that loss of toxicity of DDT on limewash is due mainly to absorption and not to any chemical change. That toxicity is lost on limewashed glass plates where absorption cannot be very deep suggests that the available layer is a very thin one.

TABLE XV.

Mean daily temperatures and humidities recorded in laboratory during period of tests.

Week ending	Mean daily temp. °F.		Mean daily relative humidity		
	Maximum	Minimum	6 a.m.	12 noon	6 p.m.
May 11th	78·7	73·1	81·5	80·4	76·6
18th	80·3	73·1	82·0	80·0	76·1
25th	79·6	74·3	81·7	82·9	76·1
June 1st	77·3	71·6	81·1	82·1	78·0
8th	78·4	72·3	80·0	79·9	75·9
15th	76·6	71·0	82·4	81·9	79·9
22nd	76·8	71·4	80·6	87·1	77·1
29th	76·6	72·4	80·6	80·9	77·4
July 6th	78·0	72·8	77·9	75·3	75·4
13th	77·4	72·7	79·6	79·0	72·3

Conclusions.

There would appear to be little doubt that considerable absorption occurs when oil solutions and emulsions are applied to vegetation and to the mud and plaster of dwellings, with a consequent loss of available insecticide. There is less loss of dispersible powders applied to such surfaces.

It is suggested that attention be directed to the production, for experimental purposes in the first instance, of dispersible powders of various particle sizes and with higher insecticide content, and perhaps of emulsions with varying viscosities and stickiness.

APPENDIX.

[The further information contained below was received from the authors just as the foregoing article was going through the press, and it has, therefore, been incorporated as an appendix.—Ed.]

DDT Dispersible Powders and Limewash.

1. Neocid B.A. 50, manufactured by Geigy Co. New York, is a finely divided homogeneous white dispersible powder containing 53 per cent. hydrolysable chloride and 38 per cent. pp′ DDT. Mixed with water to give a 5 per cent. suspension of DDT the particles fall out of suspension but on shaking mix again easily. No blockages of the nozzle of a Four Oaks "Kent" Sprayer occurred.

Glass plates limewashed two weeks previously were sprayed with a suspension of Neocid B.A. 50 containing 5 per cent. DDT in water, and 20 *G. palpalis*, one week old, were exposed for 15 seconds contact to each plate two days and two weeks after the application.

Percentage mortalities and the amounts of DDT recovered from the plates were as follows :—

Treatment of glass plates	Age of deposit	Percent. kill of *palpalis* in 24 hrs.	Recovery of pp′ DDT mgms./sq. ft.
Lime + Neocid (1)	2 days	100	172
,, ,, ...	2 weeks	100	
Lime + Neocid (2)	2 days	100	83
,, ,, ...	2 weeks	100	
Neocid only (1)	2 days	100	63
,, ,, ...	2 weeks	100	
Neocid only (2)	2 days	100	95
,, ,, ...	2 weeks	100	

2. N.210 Spray Powder, containing 19·6 per cent. hydrolysable chloride and 14·3 per cent. pp′ DDT, is manufactured by Hygienic Chemical Co. Mixed with water the larger particles rapidly fall out of suspension, leaving a dark brown liquid above a whitish deposit of various sized particles.

Limewashed glass plates were sprayed with a suspension of N.210 spray powder in water containing 2 per cent. DDT, and 20 *G. palpalis* were exposed for 15 seconds contact to each plate two days and two weeks after the application.

Percentage mortalities and amounts of pp′ DDT recovered from the plates are given below :—

Treatment of glass plates	Age of deposit	Percent. kill of *palpalis* in 24 hrs.	Recovery of pp′ DDT mgms./sq. ft.
Lime + N.210 (1)	2 days	90	42
,, ,, ...	2 weeks	95	
Lime + N.210 (2)	2 days	95	42
,, ,, ...	2 weeks	95	
N.210 only (1)	2 days	15	53
,, ,, ...	2 weeks	10	
N.210 only (2)	2 days	5	47
,, ,, ...	2 weeks	5	

These results were so surprising that the tests were repeated with the following results :—

Treatment of glass plates	Age of deposit	Percent. kill of *palpalis* in 24 hrs.	Recovery of pp′ DDT mgms./sq. ft.
Lime + N.210	24 hours	85	39
,, ,, ...	2 weeks	90	
N.210 only	24 hours	10	50
,, ,, ...	2 weeks	10	
Lime only	—	0	0

In other tests it was seen that the surface deposit obtained after spraying a water suspension of N.210 powder on mud blocks was white, whereas the deposit on a non-absorbent glass plate was khaki in colour. This suggested that the water soluble " sticker " (sulphite lye) was absorbed into the mud leaving an effective deposit on the surface, but after evaporation of the water from the non-absorbent surface the " sticker " masked the effect of the insecticidal deposit.

A water suspension of N.210 powder was therefore allowed to stand, and the brown liquid decanted. A water suspension of the resulting white sediment was sprayed on to a glass plate. Twenty-four hours later this gave a kill of 75 per cent. of *G. palpalis* exposed for 15 seconds contact.

These results confirmed that at least two other DDT dispersible powders can be effectively applied to limewash. Even at comparatively small dosages they are much more toxic than oil solutions of the insecticide.

Analysis of the lime used in these tests was as follows :—

Insoluble in 10 per cent. hydrochloric acid ...	8·6 per cent.
Soluble in 10 per cent. hydrochloric acid :	
Calcium oxide, CaO	39·8 per cent.
Calcium carbonate, $CaCO_3$	36·6 per cent.
Combined water	8·0 per cent.
Ferric oxide, Fe_2O_3	2·6 per cent.

NOTES.
1. Part of the lime is unslaked.
2. Some alumina and magnesium present but were not estimated.
3. The iron content is high but from chemical estimations of the insecticide it does not seem that any marked decomposition of the DDT was produced.
4. Qualitative tests showed chlorides and sulphates to be absent.
5. The alkalinity of 1 g. of lime was equivalent to 14·22 ml. of normal acid when titrated with phenolphthalein as indicator.

AN EXPERIMENT WITH DDT AGAINST PESTS OF STORED PRODUCTS.

By B. M. Jones, B.Sc., F.R.E.S., Lieut. (Sp.) R.N.V.R.
Medical Department of the Navy.

E.M.N.

The published information on the toxicity of DDT to insects infesting stored products in general provides little more than lists of those insects that are apparently susceptible to the toxic action. The direct application of DDT in a kerosene oil base to bagged commodities cannot, however, be introduced as a control measure, as contamination of food products may have deleterious effects on the consumers.

Pyrethrum in P.31 oil has been recognised as the best multipurpose insecticide, mainly because there is no risk of harmful effects on consumers. It is also very effective against larvae and adults of economically important moths. Pyrethrum in this form can be applied by means of an Aerosol or Freon " bomb ", which is a container of American design, weighing approximately 26 oz. and containing 18 oz. of insecticide. The insecticide consists of 0·4 per cent. pyrethrum and 6 per cent. sesame oil in " Freon " (dichlorodifluoro-methane) which acts as a propellent. The propellent is under pressure and when a small screw cap is removed from a nipple, which usually protrudes at one end of the container, the contents are emitted through a minute aperture as an atomised spray.

This insecticide is quite effective against both larvae and adults of the moth, *Corcyra cephalonica*, Staint., but is practically non-toxic to hard-cuticled insects, such as *Calandra oryzae*, L., and *Tribolium castaneum*, Hbst.

The advent of DDT was generally acclaimed for its extraordinary toxic properties, but simple tests, in which the insect was allowed to be continuously exposed to a residual deposit of DDT, showed that larvae of the moths, *Corcyra cephalonica* and *Ephestia cautella*, Wlk., which are economically important in relation to stored food, were completely resistant. In other preliminary tests of this kind, using *Calandra oryzae*, *Tribolium castaneum*, *Rhizopertha dominica*, F., and *Laemophloeus* spp. as test insects, this was not the case, for they quickly displayed the narcotic symptoms characteristic of DDT poisoning, and it appeared that such hard-cuticled insects would succumb to the residual or persistent effect of this toxicant.

Evidence from experiments on the toxicity of DDT towards warm-blooded animals indicates that concentrations of DDT within the range of 1 to 10 per cent. as used in the field are not harmful to human beings. The effect on Sinhalese coolie operators engaged on spraying operations at the R.N. School of Tropical Hygiene, Colombo, during 1945, as fully described by Stammers & Whitfield (1947), bears this out.

DDT was not used by the Services on a large scale until 1943–44 and therefore literature on its toxicity towards human beings cannot, at this stage, include any real evidence to show whether the cumulative effect of DDT may or may not prove harmful. Until such evidence is available, the policy is to avoid contamination of foodstuffs with DDT and the method of application adopted in the work described below was designed with this end in view.

As Pyrethrum and Aerosol " bombs " were not available at the time, experiments were made with DDT in order to find a method that would eliminate the risk of contaminating foodstuffs.

The purpose of this work was twofold :—

(*a*) To determine the biological activity and control value of a surface film of DDT emulsion-gum arabic solution when applied to bags.

(*b*) To assess biologically the efficiency of this film as a medium for retaining the DDT deposit as an external layer upon the bags.

Nocturnal Activity of Insects.

A method, whereby DDT could be used as a means of controlling insects attacking stored food, was devised in the light of the following observations.

An insight into the behaviour of insects in stored food was provided by examination at night when it appeared that greater numbers migrated to and over the surface of the bags than by day. Although it was not attempted, it would have been interesting to record the insect density on known surface areas throughout at least a period of 24 hours.

This increased activity at night pointed to the possibility that, should the percentage of insects migrating to the surface be sufficiently high, the application of an external layer of DDT deposit would effectively control insects established in the food commodity contained in the bags.

During storage, localised and favourable conditions of temperature, relative humidity and moisture content may be responsible for a relatively localised infestation, which during inspection is sometimes not detected. From this source of infection, the insects, *e.g. Laemophloeus* spp., migrate, causing widespread and in due course serious infestation.

Moreover, endemic insect populations in unhygienic storage buildings are responsible for most foodstuffs becoming infested. Migration of insects from the building fabric to the bagged commodities is unavoidable, if no precautionary measures are taken. Under present day conditions, food reserves are built up, usually in unsuitable storage places, that provide ideal conditions for insect multiplication. This is particularly true of the tropics, where the hot humid climate is responsible for accelerating the life-cycle.

The persistent toxic effect of a DDT deposit laid down as a protective coating upon bagged commodities would be an important factor in preventing infestation of this kind.

Experimental Work.

An emulsion concentrate "Mayonnaise cream" type Code No. T.P.521 was used, because DDT in emulsion form has been proved to be in one of its least toxic forms to warm-blooded animals.

Composition of the concentrate :—

	Per cent.
DDT Technical (60·5 per cent. pp′ isomer)	30·0
Solvent naphtha	40·01
Casein	1·45
Phenol	0·64
Agral W.E.M.	0·5
Caustic potash	0·22
Water	26·8
Ethyl alcohol	0·38

The concentrate was diluted with water and gum arabic solution to produce a 5 per cent. DDT emulsion-gum arabic solution.

The gum arabic component of this heterogeneous carrier medium gives the latter a consistency that retards the tendency of the emulsion, either by absorption or capillary action, to penetrate or "creep" through the sacking texture. The solution thus remains as a surface film. The volatile components evaporate and the gum arabic retains the deposit of DDT as an external layer.

This was the theory upon which the experiment was based, the gum arabic giving the "adhesive" quality to the carrier medium.

The 5 per cent. DDT emulsion-gum arabic solution was prepared as follows :—

Emulsion concentrate (30 per cent. DDT) ...	20 parts
Gum arabic (16 per cent. in water)	25 parts
Water	75 parts

Twenty parts of the emulsion concentrate was first diluted with 20 parts of water and thoroughly stirred. The remaining volume of water and the 25 parts of the gum arabic solution was added with additional stirring.

The introduction of the gum arabic into the heterogeneous composition of the emulsion did not impair the physical stability. The emulsion-gum arabic solution remained stable for a period of 5 hours, which is well within the specification for emulsion insecticides.

Each jute bag for the experiment had a surface area of approximately 224 sq. in. The bags were treated with an atomised spray at an estimated concentration rate of 50 mgm. DDT/sq. ft. Measurements were made by exposing to the spray, around the edge of the bag, glass slides which were weighed in stoppered bottles before and after each spraying run. The concentration rate of the DDT was then calculated from the weight of the deposit on the glass slides, by referring to the known strength of the emulsion.

The nozzle of the sprayer was not directed at or near the bag surface, but in such a position as to allow the spray to settle as a concentrate mist. This procedure, after some preliminary practice runs, was more inclined to produce a uniform deposit upon the bag surface, and moreover was a reliable method for giving the light concentration of DDT required.

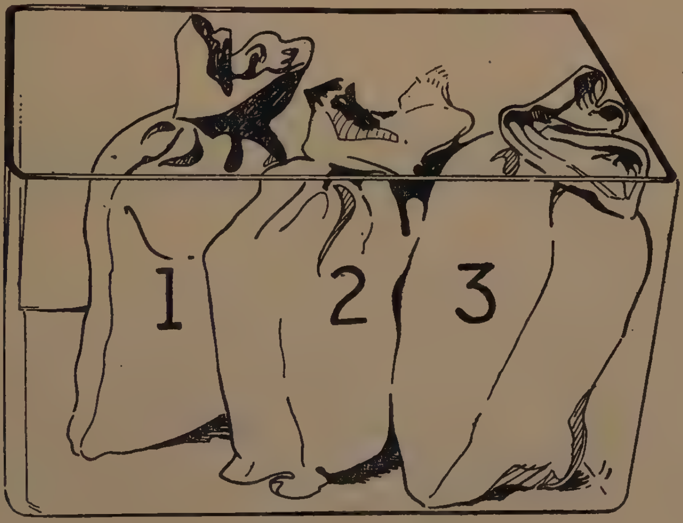

Fig. 1.

Three bags, two with treated surfaces and one without, were filled with rice and flour and placed in a glass tank, 12 in. × 9 in. × 9 in. with their opposing surfaces contiguous as shown in fig. 1. Bags Nos. 1 and 3 were filled with non-infested rice and flour, No. 1 having a treated surface and No. 3 untreated. Bag No. 2 had a treated surface and was filled with rice and flour containing 41 specimens of *Calandra oryzae*, 47 *Laemophloeus* spp., 80 *Rhizopertha dominica* and 67 *Tribolium castaneum*.

Observations were made each day at hourly intervals. The "knock down" insects were collected and transferred to clean petri dishes. Not one "knock down" insect made a recovery from the narcotic effect of the DDT and death followed in 6 to 15 hours.

A kill for the purpose of fig. 2 was defined as an insect "knocked down" by the narcotic effect of the DDT and from which there was no recovery.

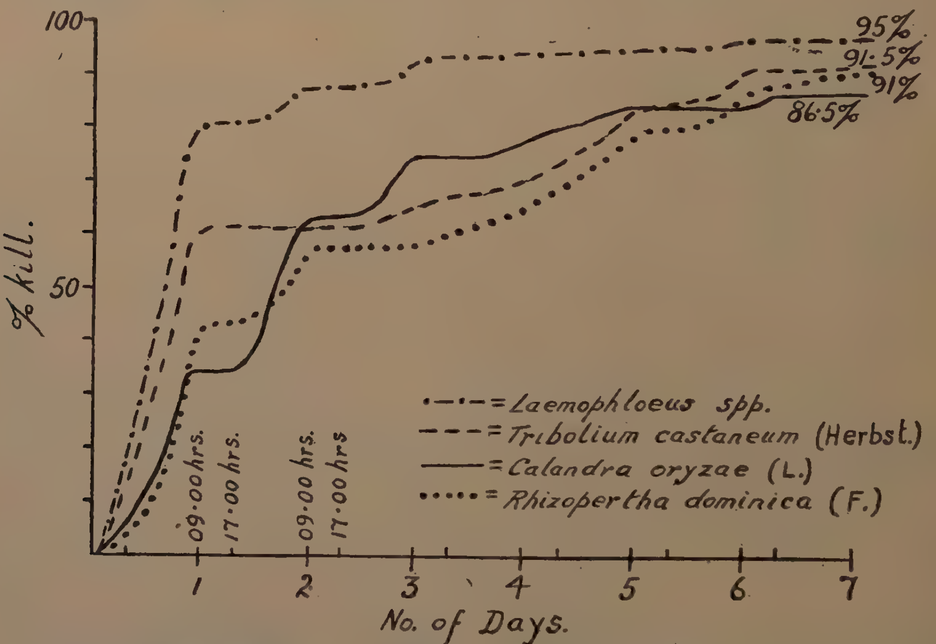

Fig. 2.—Biological effect and control value of DDT emulsion-gum arabic solution when applied as a surface film to bags of food products.

An interesting feature of the results shown in fig. 2 was a nocturnal-insect activity increase relationship indicated in the case of the four test insects.

There was little or no insect activity upon the bag surfaces during daylight and this was responsible for a relatively low kill during this period, a feature illustrated by the horizontal sections of the graph lines.

This nocturnal increased activity of the test insects, which was indirectly measured by the experiment, verifies the observations made in the field.

Of the insects contained in the bag, 60 per cent. of *Tribolium castaneum* and 80 per cent. of *Laemophloeus* spp. were killed at the end of a 24-hour period. In 48 hours an approximately 60 per cent. kill was recorded for both *C. oryzae* and *R. dominica*. After seven days the mean percentage kill was 91 per cent. and of those insects that migrated to the bag surface, 4 per cent. succeeded in crossing over to the adjacent bags, but not one of these insects survived.

Persistent toxic Effect of a DDT Film on Jute Bags.

Batches of test insects were exposed to a bag surface that had been treated with the DDT emulsion-gum arabic solution, at a concentration rate of 50 mgm. DDT/ sq. ft. For each test run, four batches of *T. castaneum*, *C. oryzae*, *Laemophloeus* spp. and *R. dominica*, respectively, were confined under separate petri dishes, for a standard time of 3 hours.

At the end of a 3-hour exposure period, each batch of 10 insects was transferred to a clean filter paper and covered with a clean petri dish, ready for observation. The test runs were carried out, at intervals of five days, over a protracted period of time.

The results presented as block diagrams in fig. 3, show at a glance the observed mortality rate of the 10 insects, up to a period of 24 hours after exposure.

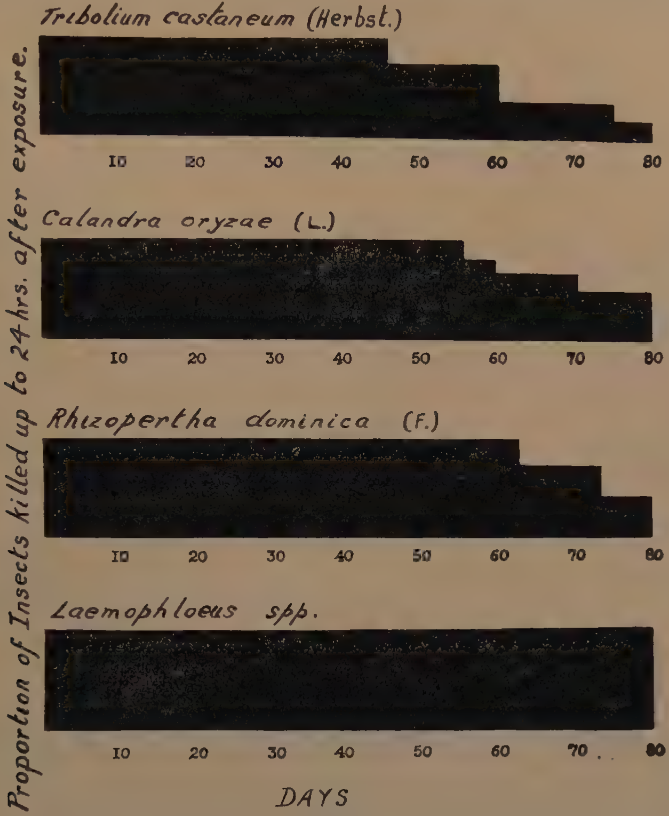

Fig. 3.

De-activation of the deposit was not shown until after the 50th day, when a lapse of more than 24 hours was required before a total kill of a batch of *T. castaneum* was recorded. About the 60th day both *C. oryzae* and *R. dominica* were also becoming more resistant within the 24-hour period of observation. *Laemophloeus* spp. appeared to be the most susceptible and a total kill was being recorded on the 80th day, when the tests were discontinued.

Biological Assessment for Retention of a DDT Film on Jute Bags.

Filter papers were placed inside bags filled to capacity with rice, so that the filter paper surfaces were contiguous to the sacking texture. The filter papers were thus relative to a peripheral layer of food pressed against the sacking texture.

The bags were sprayed with the DDT emulsion-gum arabic solution as a mist concentrate, at an estimated rate of approximately 50 to 75 mgm. DDT/sq. ft.

Filter papers were removed at intervals of 1 hour, 6 hours and 24 hours after treatment and covered with petri dishes. Mixed batches of test insects were introduced under the petri dishes and continuously exposed to the filter papers. The results are shown in Table I.

TABLE I.

Interval before removal of filter paper from the treated bag	Test insects exposed to treated filter paper		Bag A	Bag B	Bag C
	Species	No.			
1 hour	*C. oryzae* *R. dominica* *T. castaneum*	2 5 3	1 *T. castaneum* killed in 6 days	No kill up to 6 days	No kill up to 6 days
6 hours	,,	,,	No kill up to 6 days	No kill up to 6 days	2 *T. castaneum* killed in 6 days
24 hours	,,	,,	No kill up to 6 days	No kill up to 5 days	No kill up to 5 days

The biological tests indicated that the DDT did not penetrate through the sacking texture and contaminate the filter papers. Control tests were carried out by exposing insects to sprayed (at rate of 50 mgm. DDT/sq. ft.) and unsprayed filter papers. The results are shown in Table II.

TABLE II.

Filter paper surface	Insects exposed to filter papers		Result
	Species	No.	
Sprayed at the rate of 50 mgm./sq.ft. ...	*R. dominica* ... *T. castaneum* ...	3 16	Total kill in 7 hours
Unsprayed	*R. dominica* ... *T. castaneum* ...	3 16	2 *T. castaneum* dead, up to 6 days

Summary.

(1) The experiment was carried out on a short term basis due mainly to the limited time available for stored food products work, but it does show the possibilities of giving to the carrier medium an " adhesive " quality for the purpose of retaining an external layer of DDT upon bagged commodities.

(2) The increased activity at night of insects attacking stored food is interesting and the movement of insect densities in bags containing foodstuffs needs further detailed investigation. It does seem that a very high percentage of adult beetles will migrate to the surface of bags from inside the contained foodstuffs, and this makes it possible to achieve a fair measure of control by coating bags with a deposit of DDT.

(3) When a concentrate mist of a DDT emulsion-gum arabic solution is allowed to settle upon a bag surface, there is no detectable penetration of DDT through the sacking texture and therefore no danger of contaminating the contained foodstuffs.

(4) A deposit of such a DDT film, at a concentration rate of 50 mgm. DDT/sq. ft. upon a jute bag surface, remains appreciably toxic to *Tribolium castaneum*, *Calandra oryzae*, *Rhizopertha dominica* and *Laemophloeus* spp. for a period of approximately 60 days.

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AN ARSENIC-RESISTANT TICK AND ITS CONTROL WITH GAMMEXANE DIPS.

By A. B. M. WHITNALL, M.Sc. and B. BRADFORD, M.Sc.

Y.b.

(PLATES III AND IV.)

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Introduction.

Entomologists in many parts of the world have experienced the phenomenon of resistance among insects to recognised chemical insecticides. Some workers maintain that repeated applications of chemicals result in an unintentional artificial selection of those mutants within the pest population which happen to be resistant to the poison used. The progeny of these surviving individuals develop strains of the pest that are more difficult to control than the general population. Other investigators hold that the resistant strains develop suddenly as the result of a single mutant, a proportion of the progeny of which are not killed by the chemical used, and thus quickly establish themselves.

The first record of this resistance of an insect to chemical control came from the State of Washington (Melander, 1914), when it was noted that the normal lime-sulphur spray was not altogether effective against the San José scale, *Aspidiotus perniciosus*, Comst. Quayle (1916) showed that a strain of the red scale of citrus, *Aonidiella aurantii*, Mask., had been found in California to be resistant to fumigation with HCN gas. This has also been experienced in South Africa in the Kat River Valley. The problem, in relation to this particular insect, has been studied in great detail in America, and (Dickson, 1940) showed that the factor of resistance was situated in the X-chromosome and is thus sex-linked. Two other species of Coccids in California, the black scale, *Saissetia oleae*, Bern. (Quayle, 1916), and the citricola scale, *Coccus pseudomagnoliarum*, Kuw. (Quayle, 1938), showed this increased resistance to HCN gas, and the spread of the latter was rapid.

The development of resistance among insects to recognised chemical insecticides is not confined to the Coccids, but has also been found in the codling moth, *Cydia (Carpocapsa) pomonella*, L., where lead arsenate sprays are used. In Colorado, U.S.A., two applications of the insecticide were sufficient to control the moth in 1900, but by 1930, eight to twelve applications were required to effect the same degree of control (Hough, 1928, 1929, 1934). This has also been experienced by entomologists in the Western Cape Province of South Africa.

Increased resistance against tartar emetic has been noted in the citrus thrips, *Scirtothrips citri*, Moult., in California, where it was first noted in 1942. An interesting feature was that the resistant strain developed only three or four years after the first use of the bait (Boyce, Persing and Barnhart, 1942). Smith (1946) has shown that tartar emetic is effective against the South African citrus thrips, *S. aurantii*, Faure, in the Rustenburg area, and mentions the experiences of the senior author and others who have found thrips in the Kat River and Nelspruit areas to be resistant to tartar emetic. In these latter instances it appeared that the thrips had never been susceptible to the insecticide.

The primary screw worm, *Cochliomyia americana*, Cush. & Patt., has developed a resistance to phenothiazine and this resistance has been shown to be inherited (Knipling, 1942).

Thus, within the space of a few years, at least seven species of injurious insects have evolved strains that are more resistant to recognised chemical insecticides than the general populations of these pests.

There are, no doubt, other explanations for this bio-chemical development in addition to the two theories stated above, and it is hoped that this paper will throw some light upon this disturbing phenomenon. Certain experiments carried on over a period of six years with an Arachnid, the blue tick, *Boophilus decoloratus*, Koch, will be discussed as it is claimed that strains of this tick have developed a resistance to arsenic. Experiments will also be discussed which prove that this tick, and in addition certain other species, can be controlled by dips containing Gammexane.

History of the Investigations.

In view of the records detailed above, entomologists were not unduly surprised to hear in 1938 that a tick had developed a resistance to arsenic. The presence of this tick was first indicated in the East London District in 1938–39, when farmers found that they could not keep their cattle free from this one-host tick, *B. decoloratus*, by regular dippings in 7-day strength arsenical wash. Early in 1940 the observations of farmers were confirmed by the Division of Veterinary Services, for controlled dippings in various arsenical washes failed to free the cattle from these ticks (du Toit, Graf and Bekker, 1941). The Division of Veterinary Services, by field experiments, were the first to indicate that an arsenic-resistant variety or strain, morphologically indistinguishable from the blue tick *B. decoloratus*, did exist. But, as will be shown, actual proof of "resistance", as far as it has yet been possible to prove this development, was not established until later.

In July 1940, there were complaints from the East London area about the ineffectiveness of some arsenical dips. It was claimed that certain proprietary preparations were no longer effective against the blue tick. Investigations on the spot confirmed these reports, despite the fact that other species of ticks were readily killed by these arsenical washes.

It was suggested that some experiments with the resistant ticks concerned should be conducted in the Zoology Department of Rhodes University College at Grahamstown. Advice from different sources, however, indicated that any experiments should be conducted with the parasites on their hosts, as *in vitro* experiments with ticks were said to be entirely unreliable. Accordingly, the first series of experiments planned at Grahamstown was designed to include the treatment of ticks on hosts.

It was obvious that the toxic action on ticks had a direct bearing on arsenic resistance and at this stage of the investigations an attempt had to be made to solve this problem. Very little progress was made with the experiments along the lines planned, principally owing to the cumbersome technique of treating ticks on a host and to the shortage of trained workers.

In December 1940, the problem was tackled from a different angle, and Dr. J. C. van Hille of the Zoology Department of Rhodes University College undertook certain investigations. It was decided that, in spite of the warnings, *in vitro* experiments with ticks would be conducted and, in addition, a series of chemical analyses would be made of ticks treated with arsenic, while still attached to their hosts. The tick material would be fully engorged adult females. Once again the results from *in situ* tests were inconclusive, but *in vitro* immersion of presumed arsenic-sensitive ticks in 0·2 per cent. As_2O_3 indicated that arsenic is taken up by the integument of the ticks, resulting in their death. Ticks so killed did not contain arsenic in their internal anatomy. The absorption of arsenic by the integument was slow, taking about 1½ hours' immersion before its presence could be demonstrated chemically. Further, *in vitro* immersion experiments with proprietary dips and sodium arsenite seemed to indicate a superiority of the sodium arsenite over the proprietary dips, as the integument absorbed more arsenic from sodium arsenite than from the proprietary dips with the same As_2O_3 content.

These *in vitro* experiments were far removed from natural conditions, as ticks were immersed for long periods and then washed to remove excess arsenic before attempting chemical analyses. It was found that if allowed to dry on the ticks, all dips were more or less equally effective in killing adult females which were presumed arsenic-sensitive. It was obvious, then, that some standard technique should be adopted if biological observations were to be recorded from ticks treated by *in vitro* immersions.

By 1942, the indications were that the arsenic-resistant tick had established itself in the Bathurst area and as the work progressed it was felt that the *in vitro* tests gave reliable results, and the reaction of adult female ticks to treatments in the laboratory could be taken as an indication of their reactions when treated on their hosts. All the evidence that had been gained so far indicated that arsenical dips killed by contact.

Some Field Experiments.

The experiences relating to sodium arsenite and proprietary dips recorded above suggested an investigation on the wetting properties of dips, and there was also some indication that the ratio of As_2O_3 to Na_2O might have an influence on the effectiveness of the arsenic. During the period October 1943 to May 1944, the investigation was carried a stage further when field trials with three dips with ratios of arsenic to soda of 3 : 1, 5 : 1 and 7 : 1, were carried out at the " Langholm Estates ". At the same time an arsenical dip with a special wetting agent was tested at Mr. V. Webb's farm " Tharfield ". These experiments gave negative results, but they certainly confirmed the presence of an arsenic-resistant tick in the Bathurst area, for none of the dips tested effected a satisfactory control of ticks in the field.

A further field experiment using an arsenical wash in conjunction with nicotine sulphate was carried out at " Tharfield " over the period August 1944 to August 1945. This combination had been recommended by the Division of Veterinary Services (Bekker, 1945). After dipping cattle at weekly intervals for one year in this combination using 0·16 per cent. As_2O_3 and 0·04 per cent. nicotine, no ill effects were noted on the cattle, and the wash could be kept at standard strength if the farmer kept a rigid check on replenishments.

The arsenic nicotine wash was not effective against adult females of the arsenic-resistant blue tick, but cattle dipped at regular weekly intervals were kept practically free from tick life. It thus seemed that the arsenic-nicotine wash was effective against some other stage of the arsenic-resistant tick, and laboratory experiments showed the effectiveness of the wash against the larval stage.

The Advent of DDT and Gammexane.

The assistance of a whole-time worker, Miss E. M. Fenwick, in September 1944, enabled the laboratory technique to be given exhaustive tests. Attention was turned to DDT, but initial trials with this new insecticide did not meet with success.

In December 1944, a small quantity of an insecticide, then known as " 666 " became available.

Five different formulations of " 666 " were received :—

(1) Current crude product.
(2) Refined " 666," said to be four times as toxic as the crude product.
(3) 20 per cent. " 666 " with gypsum.
(4) Redispersible powder consisting of 50 per cent. crude " 666."
(5) A miscible oil containing 20 per cent. refined " 666 " in methanol with turkey red oil.

Later, Slade (1945) announced that " 666 " was hexachlorocyclohexane, of which there were four isomers known, alpha, beta, gamma and delta, but only the gamma isomer (Gammexane) was biologically active. Crude hexachlorocyclohexane contains approximately 10 to 12 per cent. gamma isomer, and it is on this basis that the percentage Gammexane has been calculated and presented in the tables.

On 18th December, 1944, " 666 " was first tested against the arsenic-resistant tick, when an experiment using the miscible oil preparation was designed to ascertain what concentration, if any, of this formulation would prove effective against adult females of this blue tick. Concentrations ranging from 0·004 per cent. to 4 per cent. refined " 666 " were tested against controls treated in turkey red oil. Twelve hours after treatment the effect of the " 666 " on the ticks was very apparent. All the ticks treated in " 666 " were flattened dorso-ventrally and markedly grooved, while the anterior and posterior ends were curved ventrally towards each other (fig. 1). The controls treated in turkey red oil were rounded, normally grooved and were moving.

Ticks had not been seen to react in this peculiar manner before and even at a dilution of 1 in 5,000 of the miscible oil preparation (0·004 per cent. " 666 ") there was a marked reduction in egg-laying, and no subsequent hatching, while the controls laid eggs that hatched normally. It was therefore decided to adopt the technique described below.

In the original experiments with these " 666 " preparations and other insecticides, some 3,000 adult female ticks were treated. Later, in more exhaustive experiments, some of which will be discussed below, 12,000 adult female ticks were treated. By using this technique it was possible to *prove* that an arsenic-resistant strain of *B. decoloratus* did exist. Furthermore, when experimental formulations of Gammexane dips were made, it was possible, by treating adult females in the manner to be described, to determine rapidly which formulations, and the dilutions of these formulations, should prove satisfactory in the field. It was also possible to check up on the gamma isomer content of a dip once it was placed in the dipping tank. As will be shown, predictions made from these laboratory tests have been amply proved by large-scale field trials. It is, therefore, felt that conclusions drawn from the results obtained by treating ticks *in vitro* are sound. The method is more especially suited for testing insecticides against the one-host blue tick *B. decoloratus*, strains of which have developed a resistance to arsenic.

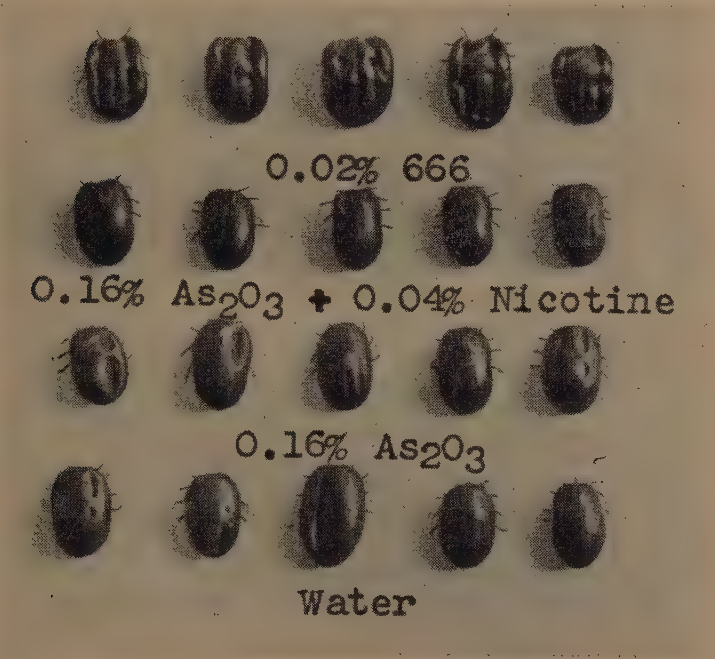

Fig. 1.—Effect of different insecticides on adult female arsenic-resistant ticks, *B. decoloratus*. The photograph was taken 24 hours after treatment, and shows the characteristic action of " 666 " on this stage of the tick.

The ticks in the top row, which were treated in 0·02 per cent. " 666 " or 0·005 per cent. Gammexane, are flattened dorso-ventrally, markedly grooved, while the anterior and posterior ends are curved ventrally towards each other.

The ticks treated in 0·16 per cent. As_2O_3 plus 0·04 per cent. nicotine and in 0·16 per cent. As_2O_3 alone, are not flattened or curved, and are normally grooved and rounded, being similar in appearance to the ticks treated in water.

Laboratory Technique.

Adult Female Tick Material.

The tick material has, in almost every instance, been taken from the " Langholm Estates," except in cases where the insecticidal susceptibility of ticks from different areas has been studied. Furthermore, it has been established by both laboratory and field tests that the blue ticks at " Langholm " were arsenic-resistant.

The ticks were removed from their hosts simply by pulling them off with the fingers. A representative sample was always taken from a number of animals in the herd. The period between the removal of the ticks and treatment varied from a few hours to three days.

Upon arrival in the laboratory the ticks were sorted into treatment groups, and placed in dishes containing filter paper. Only large, fully engorged, healthy looking adult females were selected for treatment, as experiments had shown that small, underfed specimens did not provide reliable material. At first the treatment groups consisted of 10 ticks, as this number proved satisfactory for the process of eliminating formulations. Later, when the number of formulations, or concentrations of formulations, to be tested had been reduced, a larger number of ticks, 20 or 50, were treated in the most promising formulations, and the final verdict on any preparation was based on the treatment of 100 or more ticks.

No preparatory treatment was given to the ticks as experiments had shown that sealing of the mouth-parts did not unduly affect the manner in which they reacted to the treatment.

Treatment of Adult Female Ticks in Fluids.

In the case of fluids, the method of treatment was as follows :—

The ticks were immersed for five minutes in the solutions or suspensions contained in petri dishes. After this they were removed with forceps and placed, ventral surface down, on cotton wool which had been wrung out in the same solution or suspension. Experiments had shown that a fairly wide range in the degree of dampness of the cotton wool was permissible, and it was found possible to regulate the moisture content sufficiently accurately by squeezing the cotton wool out in the hand.

The moist cotton wool was then placed on a sheet of glass, and the ticks placed ventral surface down on the cotton wool and the whole transferred at once to a fume chamber with a fan running. The ticks were left for one hour, and then placed in pairs in tubes containing a strip of blotting paper on which had been typed the relevant treatment data. The tubes were plugged with cotton wool and kept in an incubator at 25°C. and 100 per cent. R.H.

This treatment was designed to simulate natural conditions. Firstly, the ticks were completely immersed, as they would be when attached to a host passing through a dipping tank. The period of five minutes immersion was purely arbitrary. Admittedly it was longer than the time taken for an animal to pass through a dipping tank, but experiments had shown that immersion periods of 1, 5 and 10 minutes made no difference to the ultimate results of such treatments. Five minutes allowed the operator sufficient time to deal with a number of treatment groups in rotation. Secondly, the ticks were allowed to dry on moist cotton wool with a breeze playing over them. This was an attempt to simulate drying under natural conditions when the wind would dry the ticks on the hairy coat of their host. The drying of the ticks in this manner was most important for if an excessive quantity of the insecticide remained on the ticks when placed in the tubes, unreliable results would be obtained. For example, ticks known to be arsenic-resistant can readily be killed with arsenic if they are placed in too moist a condition in the tubes.

Two ticks per tube allowed individual observations to be kept on each tick and at the same time reduced the number of tubes used in each experiment. Data relating to egg-laying and hatching, such as start of laying, completion of laying, size of batch of eggs laid, start of hatching, completion of hatching and degree of hatch, were recorded for each individual tick treated. The pre-oviposition period for this blue tick under the incubator conditions of 25°C. and 100 per cent. R.H. was generally 5 to 7 days, and laying may continue for 10 days. Hatching started from 32 to 35 days after the start of egg-laying, and may continue for 17 days. These periods relate to data derived from observations after treating fully engorged female arsenic-resistant blue ticks in water, the treatment generally used as a control.

Treatment of Adult Female Ticks in Dusts.

A known quantity of the dust was placed in a test tube and the ticks inserted. The tube was gently shaken and rotated, so that the ticks were evenly covered with the dust, and the treated ticks were then immediately placed in clean tubes and likewise kept in an incubator.

Treatment of Larval Ticks.

The treatment of larval ticks in the laboratory is beset with difficulties. The larvae must be treated while housed in some container, and if a test tube containing larvae is immersed in an insecticide, the tube remains moist unduly long. Nevertheless this method gives fairly reliable results. With insecticides such as Gammexane, striking results can be obtained simply by inserting into the tubes containing the larvae strips of paper which have been moistened in the insecticide. The results obtained from the treatment of larval ticks have been expressed qualitatively, owing to the difficulty of recording counts of living larvae.

Treatment of Adult Male Ticks.

The treatment of adult male ticks *in vitro* is also difficult, for the males show a marked tendency to die shortly after removal from their hosts. For this reason laboratory experiments with male ticks have not given reliable results and they will not be discussed here, but the reaction of male ticks to Gammexane dips in the field is most striking and will be discussed later.

Insecticides tested.

Many insecticides have been tested in the search for some method to control the arsenic-resistant tick. These include different forms of arsenic ; nicotine ; preparations containing rotenone ; aloe extract plus arsenic ; " Trollope's Remedy," which includes the admixture of Little's Fluid (a disinfectant of the cresote type), dieseline and arsenic ; miscible oils ; emulsified oils ; straight oils ; proprietary cattle and sheep dips—arsenical and non-arsenical ; emulsions, suspensions and dusts containing either Gammexane or DDT.

The results obtained with some of these preparations are discussed below.

Experiments with Arsenic.

Contact Insecticidal Action of Arsenic on sensitive and resistant Strains.

Some workers maintain that the action of arsenic on the tick is purely by contact ; others that small quantities of arsenic are ingested by the tick through the peripheral blood of the host, and hence the arsenic acts as a stomach poison ; while yet others maintain that the arsenic kills by a combination of these two methods. There may be other explanations, and the habits of the one, two and three host ticks should be considered in any such discussion. The present work deals mainly with the one host tick *B. decoloratus*, and the experiments discussed below deal only with the adult females.

In the earlier experiments, in which the body contents of ticks treated in arsenic were analysed separately from the integument, the indications were that more arsenic was found in the integument than in the body contents, and the evidence seemed to favour the view that arsenic kills by contact.

This contention was further strengthened after the laboratory technique described above had been extensively tested with many different insecticides. Further, when ticks from different localities were subjected to the same arsenical treatments, and yet reacted in very different ways, it was possible to furnish actual proof that an arsenic-resistant tick did exist.

Experiments with varying concentrations of sodium arsenite, tested against presumed arsenic-resistant ticks, showed that the higher the concentration of arsenic the better was the kill of the adults. 0·16 per cent. As_2O_3 seemed to have little effect upon the adult female tick ; 0·32 per cent. As_2O_3 reduced hatching, while 0·64 per cent. As_2O_3 had a fairly marked effect but still did not give 100 per cent. control.

In another experiment using yet higher concentrations of arsenic, the results given in Table I were obtained.

TABLE I.

30 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	100	100	0
0·64 per cent. As_2O_3	73	55	60
1·28 ,, ,, ...	43	15	93
2·56 ,, ,, ...	3	100	97

This experiment indicated the degree to which these ticks could tolerate arsenic, and in subsequent experiments with arsenic, the maximum concentration used was generally 0·64 per cent. As_2O_3, which was four times the normal 0·16 per cent. As_2O_3 of 7-day wash.

A further small experiment indicated that the degree of arsenic tolerance varied greatly among ticks from different localities. An opportunity to test this hypothesis did not present itself until May 1945, when a large number of ticks, presumed arsenic-sensitive, was obtained from the Carlisle Bridge area of the Fish River. Forty-eight hours later ticks from a presumed arsenic-resistant area, Bathurst, were obtained, thus enabling 100 ticks from each area to be treated in each of the following concentrations of arsenic, 0·16 per cent. As_2O_3, 0·32 per cent. As_2O_3, and 0·64 per cent. As_2O_3. One hundred ticks from each area were treated in water as controls. The results of this experiment are given in Tables II and III.

TABLE II.

Treatment	No. of ticks commenced laying eggs in May					Total	No. of batches of eggs commenced hatching in June								Tota
	Day of month						Day of Month								
	7	8	9	10–11	12–17		8	9	10	11	12	13	14	15–25	
	Presumed arsenic-sensitive ticks—Fish River Area.														
Water ...	84	6	2	1	2	95	1	2	2	19	34	14	8	11	91
0·16% As_2O_3	6	14	8	4	5	37	0	0	0	0	0	0	1	2	3
0·32 ,,	1	2	2	0	2	7	0	0	0	0	0	0	0	0	0
0·64 ,,	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0
	Presumed arsenic-resistant ticks—Bathurst Area.														
Water ...	48	39	4	0	2	93	0	3	3	12	30	16	10	6	80
0·16% As_2O_3	20	48	19	7	1	95	0	0	1	2	7	5	12	49	76
0·32 ,,	8	44	25	3	3	83	0	0	0	1	8	10	11	38	68
0·64 ,,	3	15	23	18	4	63	0	0	0	0	7	2	4	22	35

TABLE III.

Treatment	Size of egg batch			Degree of hatching				
	Large	Medium	Small	100 per cent.	75 per cent.	50 per cent.	25 per cent.	Below 25 per cent.
Presumed arsenic-sensitive ticks—Fish River Area.								
Water	80	10	5	80	4	3	3	1
0·16 per cent. As_2O_3 ...	4	18	15	0	0	0	0	3
0·32 ,, ,, ...	0	2	5	0	0	0	0	0
0·64 ,, ,, ...	0	0	1	0	0	0	0	0
Presumed arsenic-resistant ticks—Bathurst Area.								
Water	76	16	1	64	6	7	0	3
0·16 per cent. As_2O_3 ...	54	38	3	31	29	11	1	4
0·32 ,, ,, ...	24	53	6	10	30	14	7	7
0·64 ,, ,, ...	9	33	21	5	6	12	3	9

It is felt that reliable conclusions can be drawn from these tables without resorting to statistical methods. The complete data recorded in each of the experiments can be summarised in tabular form as shown in Tables IV and V, and it is in this summarised form that the results of all further experiments will be presented.

TABLE IV.

Presumed arsenic-sensitive ticks—Fish River Area.

100 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	95	96	9
0·16 per cent. As_2O_3 ...	37	8	97
0·32 ,, ,, ...	7	0	100
0·64 ,, ,, ...	1	0	100

TABLE V.

Presumed arsenic-resistant ticks—Bathurst Area.

100 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	93	86	20
0·16 per cent. As_2O_3 ...	95	80	24
0·32 ,, ,, ...	83	82	32
0·64 ,, ,, ...	63	56	65

Ticks from the presumed arsenic-sensitive area responded readily to the arsenical treatments, as only 37 per cent. laid after treatment in 0·16 per cent. As_2O_3, while 0·32 per cent. and 0·64 per cent. As_2O_3 reduced laying to 7 and 1 per cent. respectively. The effect of the arsenic on hatching was also apparent, as only three batches of eggs laid by ticks treated in 0·16 per cent. As_2O_3 showed partial hatching. The treatments have to all intents and purposes resulted in a perfect control.

On the other hand, ticks from the presumed arsenic-resistant areas are little affected by the arsenical treatments, as laying occurred in 95, 83 and 63 per cent. of the ticks treated in 0·16, 0·32 and 0·64 per cent. As_2O_3 respectively. Hatching was also fairly heavy, as 76, 68 and 35 of the batches of eggs laid by ticks treated in 0·16, 0·32 and 0·64 per cent. As_2O_3 showed hatchings. In this case the treatments in arsenic have resulted in very imperfect controls, even with concentrations of arsenic which would injure cattle.

Another feature of these tables worthy of note, is that the arsenical solutions have produced a well marked time lag in egg-laying which is further emphasised in the hatchings.

The results of this experiment appear to furnish actual proof of the existence of an arsenic-resistant tick, and also demonstrate the marked contact insecticidal action that arsenic has on adult female arsenic-sensitive ticks.

When the 400 treated female ticks from each area had completed egg-laying, and hatching was also complete, the dead female ticks were mounted together with their progeny (Pls. III and IV). The dark backgrounds of varying intensity in Pl. III represent the many thousands of larval ticks, which were produced by the arsenic-resistant strain of *B. decoloratus*. Only one dark background appears in Pl. IV, as the arsenical treatments have been effective against the arsenic-sensitive strain. This affords a striking illustration of the problem that faces cattle farmers in arsenic-resistant tick areas.

Varying Degrees of Arsenic Resistance.

The arsenic tolerance of ticks from different localities around Grahamstown was further studied. Large numbers of ticks were obtained from Kleinemonde, the Fort Brown area on the Fish River, and Highlands. The farmers concerned had all reported difficulty in killing the blue tick with arsenical dips, and adult female ticks from these localities were treated, all about the same time, in different concentrations of arsenic. The summarised results obtained are given in the following tables; in each treatment 100 ticks were used.

TABLE VI.

Kleinemonde.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	85	95	19
0·16 per cent. As_2O_3 ...	96	88	15
0·32 " " ...	81	48	62
0·64 " " ...	19	0	100

TABLE VII.

Fort Brown.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	96	92	12
0·16 per cent. As_2O_3 ...	82	74	39
0·32 ,, ,, ...	83	75	38
0·64 ,, ,, ...	43	33	86

TABLE VIII.

Highlands.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	98	93	9
0·16 per cent. As_2O_3 ...	97	93	10
0·32 ,, ,, ...	92	73	33
0·64 ,, ,, ...	94	69	35

From Tables VI, VII and VIII, it would appear that all three farms were infected with the arsenic-resistant tick, and that the Highlands strain could tolerate more arsenic than the Fort Brown strain, which in turn could tolerate more than that from the Kleinemonde.

Decrease in Arsenic Resistance with Time, when regular Dipping is discontinued.

During 1945-46 the Eastern Cape suffered from one of the severest droughts on record and the general condition of the cattle deteriorated. In many instances the cattle were too weak to be dipped, and in any case dipping in arsenic did not alleviate the blue tick infestation. This gave an opportunity to test the influence of a period during which the cattle were not subjected to regular weekly dippings on the arsenic tolerance of ticks, and the results of these experiments (100 ticks in each treatment) are given in Tables IX and X.

TABLE IX.

Bathurst.

Treatment	May, 1945			January, 1946		
	Per cent. females laid	Per cent. batches hatched	Per cent. control	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	93	86	20	88	90	21
0·16 per cent. As_2O_3 ...	95	80	24	66	74	51
0·32 ,, ,, ...	83	82	32	21	48	90
0·64 ,, ,, ...	63	56	65	1	0	100

TABLE X.
Highlands.

Treatment	June 1945			April 1946		
	Per cent. females laid	Per cent. batches hatched	Per cent. control	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	98	93	9	100	85	15
0·16 per cent. As_2O_3 ...	97	93	10	90	80	28
0·32 ,, ,, ...	92	73	33	52	54	72
0·64 ,, ,, ...	94	69	35	20	0	100

From the above results it is seen that whereas in May and June 1945, ticks from both localities could tolerate fairly high concentrations of arsenic, in January and April 1946, ticks from the same localities could not withstand these same concentrations so readily. Once again, however, the ticks from Highlands appear to be more resistant to arsenic than those from Bathurst.

It is difficult to find an explanation for these results. They may be due to the fact that during the period when no dipping was practised there was no unintentional selection of arsenic-resistant ticks, and the number of arsenic-sensitive ticks among the population consequently increased.

It would thus appear that when once the arsenic-resistant tick is present on any farm the continued dipping of cattle in arsenic will result in the unintentional selection of a pure strain of arsenic-resistant ticks, and the degree of control must continue to decrease. On the other hand, arsenic will kill other species of ticks, as will be shown briefly later, but some other chemical is required to deal with the arsenic-resistant strain of *B. decoloratus*.

Different Forms of Arsenic.

Ticks were also treated by the *in vitro* method in the following solutions of arsenic trioxide and arsenic pentoxide.

(1) Solutions of sodium arsenite neutralised to pH 4–5 containing 0·16 per cent. As_2O_3, 0·32 per cent. As_2O_3 and 0·64 per cent. As_2O_3.

(2) Solutions of arsenic pentoxide neutralised to pH 4–5 containing As_2O_5 equivalent to 0·16 per cent. As_2O_3, 0·32 per cent. As_2O_3 and 0·64 per cent. As_2O_3.

The results are given in Table XI.

TABLE XI.
70 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	97	91	11
Sodium arsenite—			
0·16 per cent. As_2O_3 ...	51	67	66
0·32 ,, ,, ...	7	80	94
0·64 ,, ,, ...	0	0	100
Arsenic pentoxide—			
0·16 per cent. As_2O_3 ...	64	87	44
0·32 ,, ,, ...	53	81	57
0·64 ,, ,, ...	1	0	100

This experiment indicates that As_2O_3 is more effective than As_2O_5 against the arsenic-resistant blue tick, and confirms what has been known for some time, namely, that arsenates are not as toxic to ticks as arsenites (Laws, 1913, van Zyl, 1923).

Arsenic and Nicotine.

The Division of Veterinary Services (Bekker, 1945) recommended a combination of arsenic and nicotine for the arsenic-resistant tick, and these two insecticides were tested at varying concentrations, separately and combined, against adult female blue ticks from "Langholm". The results are given in Table XII.

TABLE XII.

20 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	95	100	5
As_2O_3—			
0·16 per cent.	95	84	20
0·32 ,,	100	85	15
0·64 ,,	90	83	25
1·28 ,,	35	57	80
2·56 ,,	35	29	90
Nicotine—			
0·04 per cent.	100	85	15
0·08 ,,	90	78	30
0·16 ,,	95	95	10
0·32 ,,	100	95	5
0·64 ,,	100	95	5
As_2O_3+Nicotine—			
0·16+0·04 per cent.	95	89	15
0·32+0·08 ,,	95	84	20
0·64+0·16 ,,	85	76	35
1·28+0·32 ,,	40	63	75
2·56+0·64 ,,	25	40	90

From Table XII it will be seen that once again the higher the concentration of arsenic, the greater the control exerted on the ticks. This is evident whether arsenic is used alone or in combination with nicotine. Nicotine alone has little or no effect on the ticks, and the efficiency of the arsenic is not increased by the addition of nicotine.

This experiment confirmed field observations on adult female arsenic-resistant blue ticks. But it is not meant to indicate that the arsenic-nicotine wash is not effective as a control measure for this tick. On the contrary, this wash does appear to exert a satisfactory control on the tick where regular weekly dippings are practised, and it seems likely that some other stage, larvae for example, is susceptible to the combination wash. The results from laboratory experiments with larvae have also suggested this.

The Early Experiments with Gammexane.

Miscible Oils.

Small supplies of different formulations of Gammexane arrived in South Africa in December 1944. The first preparation to be tested was the miscible oil containing 20 per cent. refined "666". This preparation mixed well with water, the solution becoming progressively more milky the higher the concentration. Dilutions ranging from 0·004 per cent. to 4 per cent. "666" (0·0015 per cent. to 1·4545 per cent. Gammexane) were tested, while as controls, ticks were immersed in solutions of turkey red oil, ranging from 0·32 per cent. oil to 16 per cent. oil. The results are given in Table XIII.

TABLE XIII.

10 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
T. R. oil—			
0·32 per cent.	100	90	10
0·8 ,, ...	100	100	0
4 ,, ...	100	100	0
16 ,, ...	100	90	10
Gammexane—			
0·0015 per cent. ...	30	0	100
0·0029 ,, ...	0	0	100
0·0073 ,, ...	0	0	100
0·0145 ,, ...	0	0	100
0·0182 ,, ...	0	0	100
0·0291 ,, ...	10	0	100
0·0364 ,, ...	0	0	100
0·0473 ,, ...	0	0	100
0·0727 ,, ...	0	0	100
0·0982 ,, ...	0	0	100
0·1455 ,, ...	0	0	100
0·2909 ,, ...	0	0	100
0·3636 ,, ...	0	0	100
0·4836 ,, ...	0	0	100
0·7273 ,, ...	0	0	100
1·4545 ,, ...	0	0	100

It will be seen above that Gammexane had a marked effect upon the adult female blue ticks, for even at a dilution of 1 in 5,000 or 0·004 per cent. "666", with a Gammexane content of 0·0015 per cent., the miscible oil gave 100 per cent. control from the treatment, while turkey red oil treatments had little or no effect.

In the second experiment the mouth-parts of the ticks were sealed with a rubber-vaseline mixture and they were then immersed in similar dilutions of Gammexane. These results are given in Table XIV.

TABLE XIV.

10 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	100	100	0
0·0015 % Gammexane	100	80	20
0·0029 ,, ...	30	10	90
0·0073 ,, ...	20	10	90
0·0145 ,, ...	0	0	100
0·0182 ,, ...	10	0	100
0·0291 ,, ...	10	0	100
0·0364 ,, ...	0	0	100
0·0727 ,, ...	0	0	100
0·1455 ,, ...	10	0	100
0·2909 ,, ...	0	0	100
0·3636 ,, ...	0	0	100
0·7273 ,, ...	0	0	100
1·4545 ,, ...	0	0	100

In general, the results expressed in Table XIV are very similar to those in Table XIII. Closing the mouth-parts did appear to affect the action of the Gammexane, particularly when dealing with low concentrations. This may indicate that small quantities of Gammexane act as a stomach poison, but this effect is not noticed when dealing with higher concentrations where the main insecticidal action is by contact. This confirmed previous experiments with other insecticides and all further experiments with Gammexane were carried out with ticks with unsealed mouth-parts.

These two experiments demonstrated the striking insecticidal action of Gammexane on adult female arsenic-resistant ticks. The characteristic flattening, grooving and curving of the ticks treated in Gammexane was noticeable in all dilutions ranging from 0·0029 to 1·4545 per cent. The ticks were not killed immediately but died off gradually, starting 9 days after treatment, and some showed leg movements up to 60 days after treatment. After 64 days all were taken as dead. The controls treated in water and turkey red oil started dying 36 days after treatment, and all were likewise taken as dead after 64 days.

Water Suspensions.

In the third experiment with Gammexane, a dispersible powder preparation containing 50 per cent. crude hexachlorocyclohexane with "Goulac" (sulphite lye) was used. When mixed with water a brown liquid was obtained, opaque in the higher concentrations, from which a cream-coloured precipitate settled. Each dilution was well-shaken before use. The results of this experiment are given in Table XV.

TABLE XV.

10 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	100	100	0
0·001 % Gammexane	30	0	100
0·002 ,, ...	30	0	100
0·005 ,, ...	20	0	100
0·01 ,, ...	10	0	100
0·013 ,, ...	10	0	100
0·02 ,, ...	0	0	100
0·025 ,, ...	10	0	100
0·034 ,, ...	0	0	100
0·05 ,, ...	0	0	100
0·067 ,, ...	0	0	100
0·1 ,, ...	0	0	100
0·2 ,, ...	0	0	100
0·25 ,, ...	0	0	100
0·334 ,, ...	0	0	100
0·5 ,, ...	0	0	100
1·0 ,, ...	0	0	100

The above results show water suspensions of Gammexane to be extremely toxic to the tick. Once again the ticks died off gradually. Those treated in water were all taken as dead 48 days after treatment, while some Gammexane-treated ticks lived longer, a few remaining alive up to 72 days after treatment. There appeared to be no relation between the strength of Gammexane and length of time elapsing before death. Ticks treated in water often die sooner on account of exhaustion after egg-laying, while those treated in effective concentrations of Gammexane are not killed rapidly, but are so affected physiologically that they cannot lay, or else lay only very small batches of eggs that are unlikely to hatch.

Dusts.

Further experiments using dusts were also carried out. A preparation containing 20 per cent. crude hexachlorocyclohexane with gypsum was diluted with kaolin to give a range of dusts containing varying percentages of Gammexane. The results are given in Table XVI.

TABLE XVI.

10 ticks in each treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Kaolin	100	100	0
0·01 % Gammexane...	100	100	0
0·02 ,, ...	100	100	0
0·05 ,, ...	60	66	60
0·1 ,, ...	10	0	100
0·2 ,, ...	0	0	100
0·5 ,, ...	20	0	100
1·0 ,, ...	10	0	100
2·0 ,, ...	0	0	100

Gammexane, when applied as a dust to adult female ticks, has the effect of suppressing laying and hatching, and also kills the ticks. At concentrations below 0·05 per cent., the insecticide has little or no effect. 0·05 per cent. Gammexane is not entirely effective, but 0·1 per cent. and higher is toxic. From the previous experiments with emulsions and suspensions the effective concentration of Gammexane appeared to be about 0·005 per cent. or even lower. It would thus appear that suspensions and emulsions of Gammexane are 20 times more toxic to the adult blue ticks than the dust preparations.

These conclusions were confirmed in another experiment using the critical dilutions of two dusts; the gypsum-crude hexachlorocyclohexane further diluted with kaolin, and the crude hexachlorocyclohexane diluted only with kaolin. The results are given in Table XVII.

TABLE XVII.

20 ticks in kaolin. 30 ticks in each Gammexane treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Kaolin	100	100	0
Crude hexachlorocyclohexane, gypsum and kaolin—			
0·02 per cent. Gammexane	100	90	10
0·05 ,, ,, ...	73	82	40
0·1 ,, ,, ...	33	9	97
Crude hexachlorocyclohexane and kaolin—			
0·02 per cent. Gammexane	100	97	3
0·05 ,, ,, ...	80	83	33
0·1 ,, ,, ...	57	41	77

The hexachlorocyclohexane-gypsum mixture gave better results, which were probably due to the more uniform mechanical preparation of this mixture as compared with the uneven mixing of the kaolin and sticky hexachlorocyclohexane which was obtained in the laboratory.

These experiments with dust again confirmed the toxicity of Gammexane to adult females of the arsenic-resistant tick. In May 1945, a small experiment was carried out in the field when three head of cattle were dusted at weekly intervals with 0·1 per cent. Gammexane dust. The dust did not harm the animals but the control of ticks was not striking. Thus no further work was undertaken with dusts, as such preparations were considered unsuitable for the treatment of cattle in the field.

Gammexane compared with other Insecticides.

An experiment to compare the toxicities of Gammexane, DDT, arsenite of soda, rotenone and nicotine was conducted. The results of this experiment are given in Table XVIII.

TABLE XVIII.

50 ticks in water. 10 ticks in each insecticidal treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	100	100	0
0·0004 per cent. Gammexane	80	100	20
0·0007 „ „	80	63	50
0·0015 „ „	60	50	70
0·0029 „ „	30	0	100
0·0073 „ „	0	0	100
0·0145 „ „	0	0	100
0·0291 „ „	0	0	100
0·0727 „ „	0	0	100
0·1455 „ „	0	0	100
0·0025 per cent. DDT (Technical) ...	100	90	10
0·005 „ „ „ ...	100	90	10
0·01 „ „ „ ...	100	100	0
0·02 „ „ „ ...	100	100	0
0·05 „ „ „ ...	100	100	0
0·1 „ „ „ ...	100	90	10
0·2 „ „ „ ...	100	90	10
0·4 „ „ „ ...	70	86	40
1·0 „ „ „ ...	60	66	60
0·16 per cent. As_2O_3	100	60	40
0·32 „ „	100	60	40
0·64 „ „	60	0	100
1·28 „ „	40	0	100
0·00125 per cent. Rotenone	100	100	0
0·0025 „ „	100	100	0
0·005 „ „	100	80	20
0·01 „ „	100	100	0
0·0125 „ „	100	100	0
0·025 „ „	90	100	10
0·05 „ „	100	80	20
0·1 „ „	100	90	10
0·01 per cent. Nicotine	100	80	20
0·02 „ „	100	100	0
0·04 „ „	100	80	20
0·05 „ „	100	80	20
0·1 „ „	100	90	10
0·2 „ „	100	100	0
0·4 „ „	100	100	0
0·8 „ „	100	100	0

Once again the toxicity of Gammexane to adult females of the arsenic-resistant tick was demonstrated, and it proved to be the most effective insecticide tested in this experiment, bringing about complete control at a dilution of 0·0029 per cent. Gammexane, while treatment in 1 per cent. DDT resulted in only 60 per cent. control.

In yet another experiment, Gammexane was compared with other insecticides which were claimed to have tick-killing properties. An emulsion was made up by dissolving 10 gms. of refined hexachlorocyclohexane, containing approximately 30 per cent gamma isomer, in 58 gms. of methyl alcohol and then shaking up with 132 gms. of turkey red oil. This 5 per cent. emulsion was diluted with water to give a range of Gammexane concentrations which were tested alongside varying concentrations of the original miscible oil. At the same time anabasine sulphate, sodium arsenite, and the so-called "Trollope's Remedy" were tested. This latter "remedy" consists of adding to every 400 gallons of 0·16 per cent. As_2O_3 wash, 1 gallon of Little's Fluid (a disinfectant of the creosote type) and 2 gallons of dieseline. These ingredients were tested separately and combined. The results of the experiment are given in Table XIX.

TABLE XIX.

50 ticks in water. 20 ticks in turkey red oil. 10 ticks in each insecticidal treatment.

Treatment	Per cent. females laid	Per cent. batches hatched	Per cent. control
Water	100	94	6
0·08 per cent. Turkey Red Oil ...	100	100	0
4 ,, ,, ,, ...	100	100	0
Original miscible oil—			
0·0004 per cent. Gammexane ...	90	100	10
0·0007 ,, ,, ...	70	71	50
0·0015 ,, ,, ...	60	66	60
0·0029 ,, ,, ...	30	33	90
0·0073 ,, ,, ...	20	0	100
0·0145 ,, ,, ...	40	0	100
0·0291 ,, ,, ...	20	0	100
0·0727 ,, ,, ...	10	0	100
Laboratory emulsion—			
0·0004 per cent. Gammexane ...	100	90	10
0·0007 ,, ,, ...	70	86	40
0·0015 ,, ,, ...	30	0	100
0·0029 ,, ,, ...	10	0	100
0·0073 ,, ,, ...	10	0	100
0·0145 ,, ,, ...	0	0	100
0·0291 ,, ,, ...	0	0	100
0·0727 ,, ,, ...	0	0	100
0·1 per cent. Anabasine	100	100	0
0·2 ,, ,, ...	100	90	10
0·4 ,, ,, ...	100	100	0
0·8 ,, ,, ...	100	100	0
0·16 per cent. As_2O_3	100	100	0
0·32 ,, ,, ...	100	60	40
0·64 ,, ,, ...	90	22	80
1·28 ,, ,, ...	60	33	80
2·56 ,, ,, ...	40	0	100
Little's 0·25 per cent.	100	100	0
Dieseline 0·5 per cent.	80	13	90
"Trollope's Remedy"	90	44	60

The toxicity of Gammexane to ticks was again demonstrated and the minimum effective gamma isomer content seemed to range around 0·0029 and 0·0073 per cent. for the original miscible oil, and 0·0007 and 0·0015 per cent. for the laboratory emulsion.

Anabasine at the concentrations used proved useless, while the degree to which these ticks could tolerate arsenic was also demonstrated. Treatment in " Trollope's Remedy " resulted in 60 per cent. control. This was apparently due to the dieseline, as 0·16 per cent. As_2O_3 and Little's Fluid separately did not prevent or reduce laying or hatching, while treatment in 0·5 per cent. dieseline alone resulted in 90 per cent. control.

Reviewing the results of the experiments with Gammexane that have been discussed so far, it is evident that this new insecticide is extremely toxic to adult female arsenic-resistant ticks. While the minimum percentage necessary to kill ticks cannot be determined from these experiments, it would appear that 0·005 per cent., or a dilution of 1 in 20,000, could be taken as a basis for further experiments.

None of the Gammexane preparations tested so far was considered suitable for use as cattle dips but progress is being made with formulations that can be satisfactorily used in dipping tanks.

Summary.

During the past 30 years, at least seven species of injurious insects are known to have evolved strains that are more difficult to kill with recognised chemical insecticides than are the normal populations of these pests.

In 1939 it was reported from the East London area that farmers were experiencing difficulty in controlling the one host blue tick, *B. decoloratus*, by regular dipping in the recognised 7-day strength arsenical wash. Field observations confirmed this.

A technique for treating ticks in the laboratory is discussed in detail. This technique has been put to exhaustive test and some 15,000 adult female blue ticks have been subjected to *in vitro* treatments with remarkably consistent results.

Experiments are discussed that suggest that arsenic kills ticks by contact. It is further claimed that by using the technique described it has been possible to furnish proof that an arsenic-resistant strain of *B. decoloratus* does exist in South Africa. It would seem that when once the arsenic-resistant tick has established itself on any farm, the continued dipping of cattle in arsenical washes results in an unintentional artificial selection of those varieties within the tick population that are resistant to arsenic. Continued dippings in arsenic thus aggravate the position and some insecticide other than arsenic should be used to combat this arsenic-resistant strain of *B. decoloratus*.

Gammexane was experimented with and it is shown that 0·005 per cent. is sufficient to kill adult females of the arsenic-resistant tick and could be taken as a basis for further experiments. Oil base preparations and water suspensions of Gammexane are twenty times more toxic to the tick than are dust preparations.

Acknowledgements.

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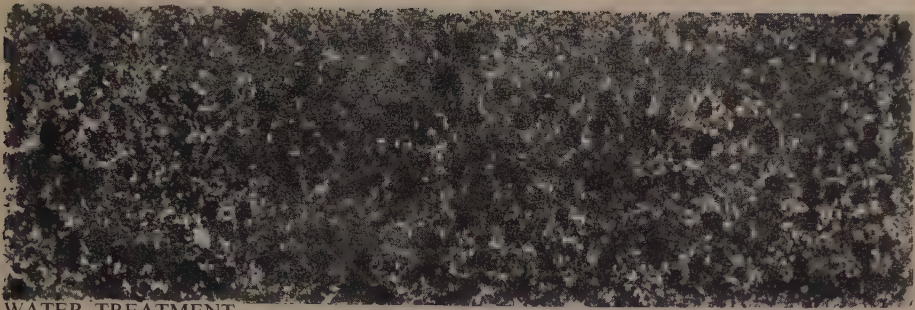
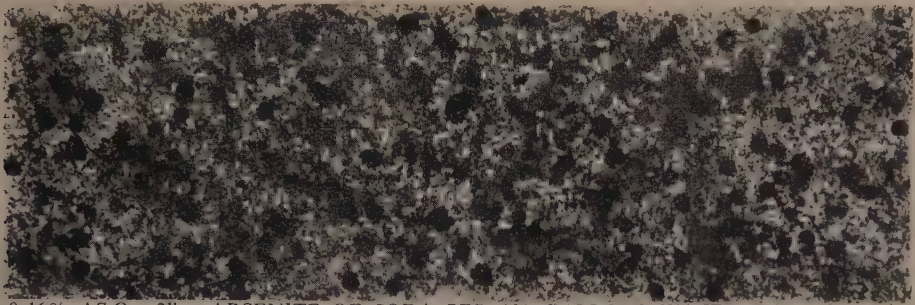
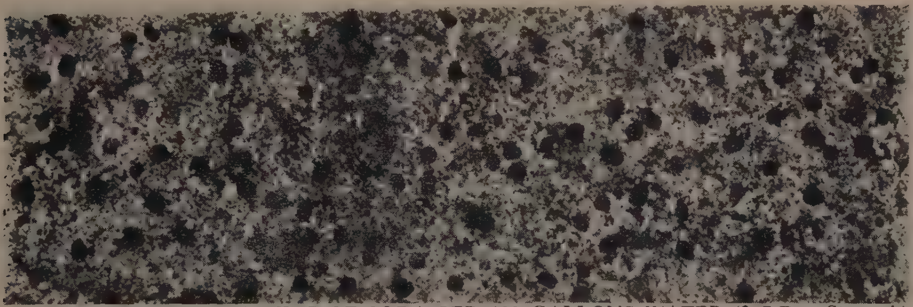


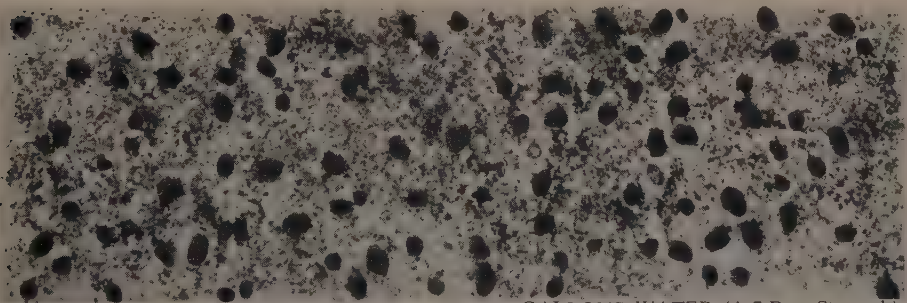
WATER TREATMENT.
Out of 100 female ticks treated, 93 laid 144,390 eggs, from which 118,308 larvae hatched.



0·16% AS_2O_3—2lbs. ARSENITE OF SODA PER 100 GALLONS WATER (7-Day Strength)
Out of 100 female ticks treated, 95 laid 129,310 eggs, from which 87,151 larvae hatched.

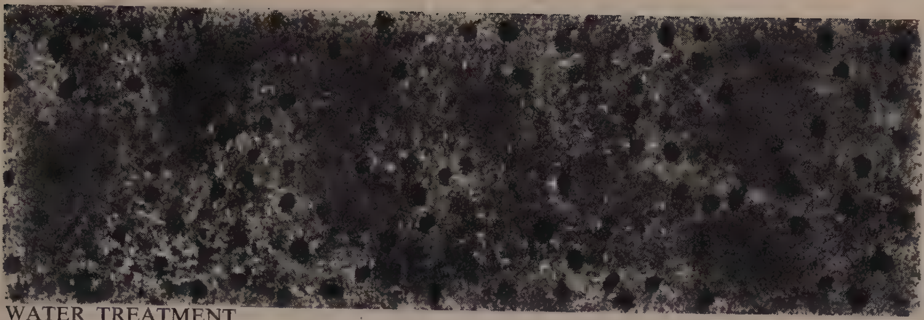


0·32% AS_2O_3—4lbs. ARSENITE OF SODA PER 100 GALLONS WATER (2x7-Day Strength)
Out of 100 female ticks treated, 83 laid 94,120 eggs, from which 50,231 larvae hatched.

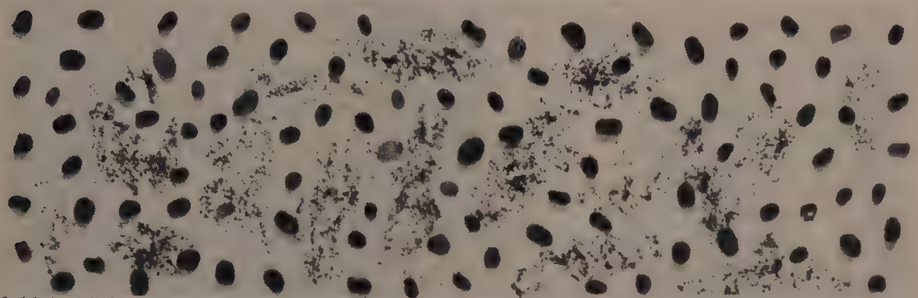


0·64% AS_2O_3—8lbs. ARSENITE OF SODA PER 100 GALLONS WATER (4x7-Day Strength)
Out of 100 female ticks treated, 63 laid 53.221 eggs, from which 21.343 larvae hatched.

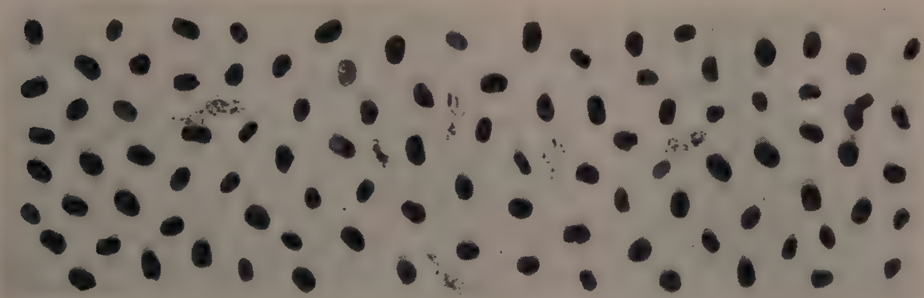
ARSENIC RESISTANT TICKS — BATHURST AREA
To show the relative ineffectiveness of arsenic when used against these ticks.



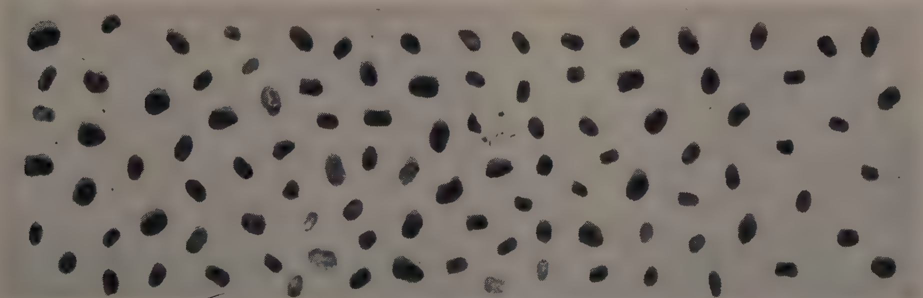
WATER TREATMENT
Out of 100 female ticks treated, 95 laid 146,350 eggs, from which 133,778 larvae hatched.



0·16% AS_2O_3—2lbs. ARSENITE OF SODA PER 100 GALLONS WATER (7-Day Strength)
Out of 100 female ticks treated, only 37 laid 28,450 eggs, from which 1,269 larvae hatched. These soon died.



0·32% AS_2O_3—4lbs. ARSENITE OF SODA PER 100 GALLONS WATER (2x7-Day Strength)
Out of 100 female ticks treated, only 7 laid 3,290 eggs, and NO larvae hatched.



0·64% AS_2O_3—8lbs. ARSENITE OF SODA PER 100 GALLONS WATER (4x7-Day Strength)
Out of 100 female ticks treated, only 1 laid 270 eggs, and NO larvae hatched.

ARSENIC SENSITIVE TICKS — FISH RIVER AREA

To show the complete control obtained when arsenic is used against these ticks.

THE BIOLOGY OF THE PARASITES OF *LOXOSTEGE STICTICALIS*, L., IN NORTH AMERICA—*METEORUS LOXOSTEGEI*, VIER. (BRACONIDAE, METEORINAE).

By F. J. Simmonds. E.M.N.
Imperial Parasite Service.

Introduction.

Meteorus loxostegei, Vier., is another important endoparasite of *Loxostege sticticalis*, L. It is perhaps second to *Bracon vulgaris*, Cress., in general importance as a control agent ; it appears to be somewhat more variable in numbers than the latter species, but neither in the present work has been seen to reach the degree of parasitism shown by *Phytodietus* in Alberta in 1942.

This species has been recorded only from North America, as a parasite of *Loxostege commixtalis*, Wlk. (Hoerner 1933), *L. sticticalis*, L. (Strickland 1921), *Phlyctaenia tertialis*, Guen. (Balduf 1929), *Pyrausta futilalis*, Led., *P. nubilalis*, Hb. (Muesebeck 1923), *P. penitalis*, Grote (Poos 1927) and *L. sticticalis*, L. (Simmonds 1945, 1947b) and is apparently of wide distribution from Massachusetts to New Mexico, Colorado and Alberta. The life-history was mentioned by Balduf who bred the species on *Phlyctaenia*. However, a number of his statements are at variance with the observations made here. He states that " *M. loxostegei* passes the winter at Urbana as a larva, presumably of advanced size, in its host ". Here the overwintering parasite larva within the host is always in the first stage. Also he states " Whenever a post-mortem (of the host) was made the exit hole of the parasite was located on the seventh abdominal segment behind the third pair of pro-legs." Fifty *Loxostege* larvae, from which *Meteorus* had emerged, were examined ; twenty-seven of these had the exit hole on the seventh abdominal segment, twenty on the eighth, and three on the ninth. In the present work unfertilised parasite eggs produced males, fertilised eggs females, and the species was not thelytokously parthenogenetic, as suggested by Balduf.

Systematic Position and Adult Characters.

Meteorus Haliday (=*Protelus* Foerster, = *Zemiotes* Foerster,=*Perilitus* Foerster (nec Nees),=*Saprotichus* Holmgren) is the sole well-defined genus of the Braconid sub-family Meteorinae monographed by Muesebeck (1923), who gives a detailed description of the generic characters from which the following short description has been taken :—Head transverse, maxillary palpi, 6-segmented ; labial 3 segmented ; eyes moderate to large, always sparsely pubescent ; mesonotum with parapsidal grooves, these usually ending in a rather large, slightly sunken, roughened area posteriorly ; disk of scutellum more or less convex ; propodeum not regularly areolated, inner spur of posterior tibiae never more than half as long as metatarsus, usually much shorter ; second cubital cell complete and subtrapezoidal, narrowed toward radius ; radius made up of three abscissae, the first two short, occasionally of equal length, but the first usually shorter than second ; abdomen ovate or lanceolate, and petiolate, very rarely sub-petiolate ; spiracles of first segment placed at or very near the middle, more often a little behind than before ; first tergite usually sculptured, and in some species provided with two conspicuous fossae (called " tracheal grooves " by Marshall) anterior to the spiracles ; remainder of abdomen smooth and polished.

Most members of the genus are internal parasites of Lepidopterous larvae but there are a few doubtful records of species attacking larvae of fungivorous Coleoptera. The cocoon is characteristic, and in most species hangs from twigs, etc., by a fine thread often several inches long. With *Meteorus loxostegei* parasitising *Loxostege*, the cocoon is formed within that of the host and such a mode of suspension does not occur.

The species under investigation here was described by Viereck (1911) as *Meteorus loxostegei* from material from *L. sticticalis*, L., taken at Rocky Ford, Colorado. This is the only name under which the species has been described.

General Biology.

As a parasite of *Loxostege*, *Meteorus* passes the winter as a first-stage larva within the hibernating host larva. Further development only occurs when the diapause of the host comes to an end, and the adult parasites emerge about the same time as moths from unparasitised larvae. Males emerge slightly earlier than females, and from part of the *Loxostege* material obtained in the autumn of 1945, 132 males and 210 females of *Meteorus* were obtained, giving 61·4 per cent. females. Material bred in the laboratory from mated females gave 58·3 per cent. females.

On emergence the females were placed singly with the males in a small cage (4½ in. × 4½ in. × 2½ in.). The males showed immediate interest in the females and as each was placed in the cage the males started moving more quickly with their wings vibrating slightly at an angle of 45° from the body. When near the female the male begins to follow her stealthily while she moves away. Finally the female stops, and the male repeatedly butts the tip of her abdomen from behind with his head, his antennae not stroking the female but lying bent over his back. After this butting has continued for a few minutes the male slowly climbs on to the back of the female and copulation ensues; this may last for ten minutes. Once mated, females resist all attentions of the males.

Mated or unmated females oviposit readily in small *Loxostege* larvae. Single female *Meteorus* placed in oviposition cages similar to those used for *Bracon vulgaris* (Simmonds 1947a) attacked *Loxostege* larvae readily. All sizes of larvae were attacked but the smaller stages were more suitable, since the large larvae were active and were too strong for the parasite. The method of attack is peculiar, and is similar to that noted by Strickland (1923) for *Meteorus dimidiatus*, Cress. The female approaches the host larva and stands motionless a little way from its side with the ovipositor bent under and forwards so that it projects under the head between the fore-legs. The female then moves as though getting the right balance on her legs, and " aims " the ovipositor at the larva. There may be one or two preliminary tentative thrusts with the ovipositor towards the larva, and finally it is thrust forward fully, the host larva is stung and an egg is laid. The difference between the tentative thrusts and a successful oviposition is soon apparent after a few of each have been seen. Following a successful oviposition the female relinquishes her peculiar stance and often cleans her ovipositor with the tibial spurs on the hind legs. During actual oviposition the ovipositor is thrust forward between the front legs to an amazing extent, the whole abdomen from directly behind the thorax being bent downwards and forwards.

Kept in the small cages in the laboratory at 75°F. and fed on raisins, honey-water, sugar and water, the average length of life of the males was 40·8 days, and females up to 62 days. Two females were each offered as many hosts as each would attack each day. One attacked 316 during her life, the other 239. Subsequent dissection showed that about 80 per cent. of these attacks resulted in oviposition indicating approximately 240 and 200 eggs laid respectively.

Description of the Immature Stages.

(*a*) *The Egg.*

At oviposition the egg is elongately ovoid, about ·20 mm. long and ·08 mm. in greatest width, with a distinct petiole (fig. 1*a*), and without apparent sculpturing. It is very small and enlarges before eclosion as has been noted for *M. dimidiatus* by Strickland (1923), although it was not found to swell rapidly when placed in water or host body-fluid as was observed in the latter species. The exact method by which this enlargement occurs has not been ascertained, but has been discussed in the case of *Bracon vulgaris* (Simmonds 1947a). In the case of *Meteorus* the enlarged egg had no tracheal supply from the host. At 75°F. the egg hatches in about 6 days when it is irregularly spherical, approximately twenty times the size on oviposition.

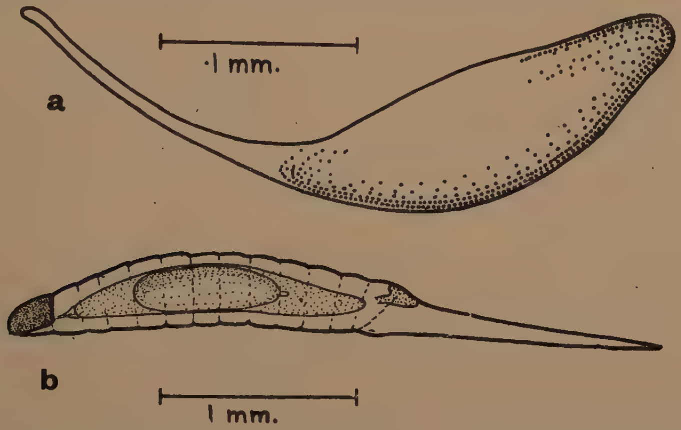


Fig. 1.—*Meteorus loxostegei,* Vier. *a,* egg at time of oviposition ; *b,* first-stage larva.

(*b*). *First-stage Larva.*

On hatching the first-stage larva is about 3 mm. in length inclusive of a long caudal appendage. Its general shape is shown in fig. 1*b*, and there appear to be 12 segments together with the head and " tail ", anterior to which the position of the closed anus is clearly visible. The tracheal system is very poorly developed ; minute lateral trunks are apparent anteriorly, but there appear to be no open spiracles. The head capsule is of the common Braconid type in which the ventral side is little developed and the dorsal and lateral parts form a chitinised shield. The mandibles are well developed, the whole structure being shown in fig. 2.

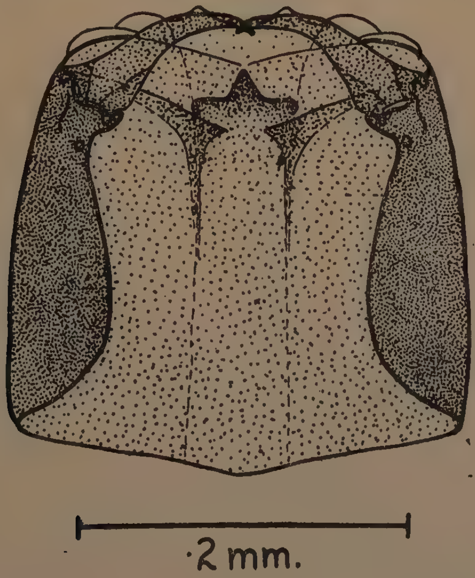


Fig. 2.—*Meteorus loxostegei,* Vier. First-stage larva, head capsule, ventral aspect.

(*c*) *Second-stage Larva.*

As mentioned above the first-stage larva does not moult until the changes prior to pupation of the host begin. The second-stage larva of the parasite is then formed (fig. 3*a*). As with *Bracon vulgaris* this is the stage of very rapid growth, and peculiar distinguishing characters are wanting. The larva is approximately 6 mm. in length and 1 mm. in width ; the head capsule is not of the chitinised type found in the first stage, and the only chitinisation is a very light rim on the mandibles, which are small and set wide apart as in the third stage. The caudal appendage of the first stage remains, but degenerates as the second stage grows. No spiracles are present, though the tracheal system is more developed than in the first stage. The second stage is of short duration, about one day, and unlike in *Bracon* it does not emerge to the exterior of the host, but changes to the third stage within the *Loxostege* larva.

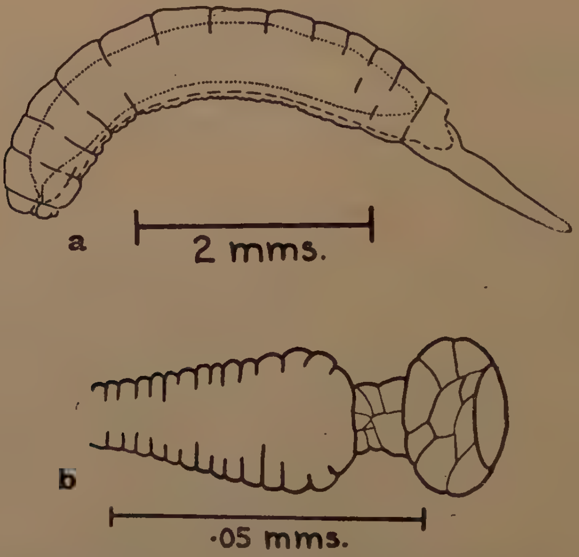


Fig. 3.—*Meteorus loxostegei*, Vier. *a*, second-stage larva, lateral view ; *b*, full-grown larva, spiracle.

(*d*) *Third-stage Larva.*

The full-grown larva is of the usual grub-like fleshy type found in the Ichneumonidae with the caudal appendage reduced to a small " hump " at the posterior end. It is about 6 mm. in length and about 1·5 mm. in greatest width, with a well developed spiracular and tracheal system (fig. 3*b*). The mouth parts and associated structures are shown in fig. 4, which shows the position of the sensoria and the small size of the mandibles, and the spinose skin is shown in fig. 5*a*.

When the larva has finished feeding it bores out through the host skin, as stated above, usually making its exit on the lateral part of the segments of the host bearing the second or third pair of pro-legs.

It is of interest to note that when parasitised by *Meteorus*, host larvae do not usually attain the same size as full-grown unparasitised larvae. This stunting of parasitised larvae also occurs with other Braconid species, *e.g. Chelonus, Ascogaster.* When the *Meteorus* larva has emerged from the host the latter is not devoured entirely or killed immediately but remains for some time alive in the cocoon, eventually dying or producing further parasites (see below).

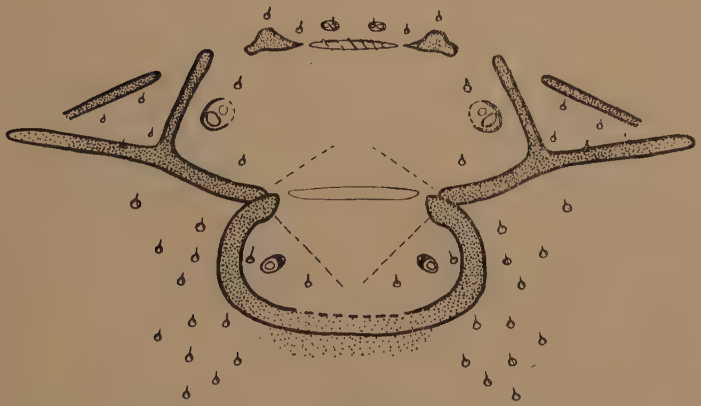


Fig. 4.—*Meteorus loxostegei*, Vier. Third-stage larva, mouth parts and associated structures.

(*e*) *The Cocoon, Prepupa and Pupa.*

Within the host spin-up the larva forms its own cocoon, which is similar to that of other METEORINAE, but lacks the suspending thread usually found in the group. The cocoon is about 5–6 mm. in length and is shown in fig. 5*b*. It is parchment-like in consistency with the surface covered with loose short silken threads. At the anterior end is a small white knob. On emergence the adult parasite bites very uniformly through the cocoon, cutting off the anterior end as is shown in the figure.

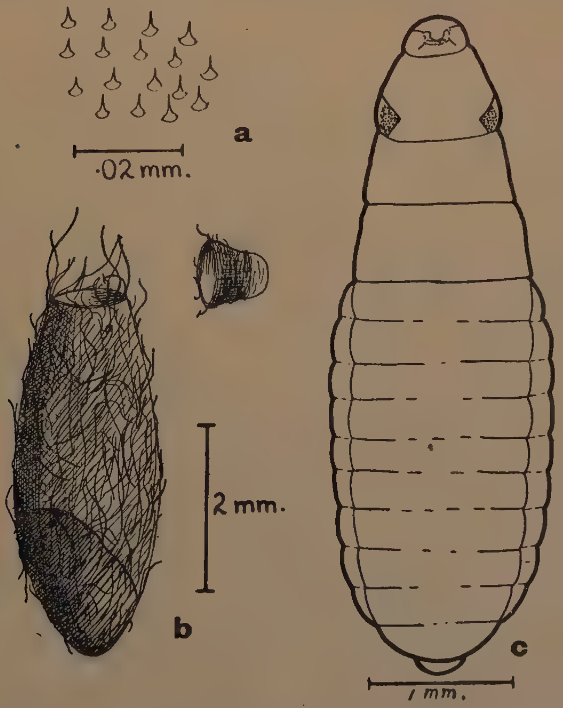


Fig. 5.—*Meteorus loxostegei*, Vier. *a*, Spinose skin of third-stage larva ; *b*, cocoon, showing mode of exit of adult by cutting off anterior end ; *c*, prepupa, ventral aspect.

Within the cocoon the larva changes to a prepupa (fig. 5*c*) which, in approximately 2 days, forms the pupa (fig. 6).

As stated above, the developmental time of the parasite is closely correlated with the behaviour of the host larva. Whatever the size of the host larva when it is parasitised, the parasite within develops to the full-grown first larval stage and remains thus without further development until the onset of changes in the host that occur prior to pupation. Thus, if the host goes into diapause the parasite remains as a first-stage larva until this diapause is broken. However, at 75°F., with hosts that pupate without arrested development, the parasite has a developmental period prior to cocoon formation of approximately ten days and about eight days subsequent to this.

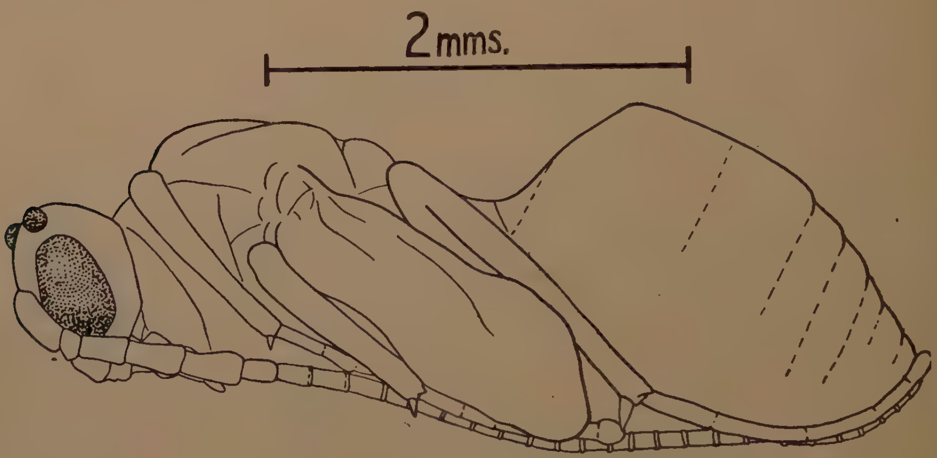


Fig. 6.—*Meteorus loxostegei*, Vier. Male pupa, lateral aspect.

Importance.

Meteorus was not present in hibernating *Loxostege* material collected in Montana in 1941, though over 2,000 hosts emerged. In 1942 it was present to the extent of 0·32 per cent. of the total living material collected. In 1945, however, it ranked second to *Bracon vulgaris* in abundance forming 3·5–4·0 per cent. of the total material collected and reaching a parasitism of 87·8 per cent. of the hibernating larvae in one field. Both *Mesochorus perniciosus*, Vier., and *Perilampus similis*, Cwfd., have been reared as secondaries from *Meteorus* cocoons, the former species in only four instances, the latter to the extent of approximately 6·2 per cent. of the total *Meteorus*. Thus these secondaries act only as a very slight check to the numerical increase of *Meteorus*. From the reproductive capacity of the females and the apparent hardihood of the species under laboratory conditions it would appear that the species is capable of higher parasitism than has been seen to occur in general in these collections. However, such deductions as to the behaviour of parasites in the field from observations made in the laboratory are often found to be worthless. In the laboratory it was found too that *Meteorus* was intrinsically superior to *Bracon vulgaris*, *Pristomerus appalachianus* var. *dorsocastaneus*, Vier., and *Campoplex sulcatellus*, Vier., and it was also found that successful multi-parasitism is often possible when *Meteorus* is one of the species involved.

This is due to the speed of development of the *Meteorus* larva once the onset of the first stage of pupation of the host initiates further growth of the parasite larva. *Meteorus* thus develops faster than other species in competition for nutritive material of the host. The *Meteorus* larva emerges from the host while the latter is still alive and still containing considerable nutritive material on which it is often possible for a second parasite species to develop, if attack has occurred previously while the *Loxostege* larva was still feeding.

Species that have been found to develop successfully in a host after a *Meteorus* larva has emerged include *Achaetoneura samiae*, Webb, *Euphorocera omissa*, Rph., /ein and the external parasite *Cryptus inornatus*, Pratt.

Since *Meteorus* seemed to be a promising parasite for introduction into South Africa as a biological control agent, attempts were made to devise a technique for breeding it in large numbers in the laboratory. However, it was found that despite numerous precautions the host was susceptible to a disease that made large scale breeding of *Meteorus* impossible. As has been described previously (Simmonds 1945), 33 75 larvae of both *Ephestia kühniella*, Zell., and *Carpocapsa pomonella*, L., were used as hosts. They were more readily attacked when coated with body fluids of *Loxostege* or *Pyrausta nubilalis*, Hb., but mortality of the parasite was high ; however, this is of interest since by this method stocks of the parasite can be maintained when *Loxostege* material is not available.

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A NEW GENUS AND SPECIES, *EURYMYIOCNEMA APHELINOIDES* (HYMENOPTERA, APHELINIDAE), AND A HISTORY OF THE GENERA *EURYISCHIA* RILEY AND *MYIOCNEMA* ASHMEAD.*

By Harold Compere.

Associate in the California Agricultural Experiment Station.

E.H.N

The new species described in this paper was received from Richard Le Pelley, Government Entomologist, Kenya Colony, British East Africa, for identification. A new genus *Eurymyiocnema* is erected for its reception. This genus is most closely related to *Myiocnema* Ashmead, which was placed by its author in the "APHELININAE", although subsequent authors have regarded *Myiocnema* as belonging to the ELASMIDAE. *Eurymyiocnema* is placed in the APHELINIDAE, and the opinion is expressed that *Myiocnema*, as well as *Euryischia* Riley, are out of place in the ELASMIDAE, since both of these genera resemble *Aphelinus* Dalman much more closely than they do *Elasmus* Westwood.

So far as is known, the species of the genera under consideration here are all secondary parasites and injurious. The species of *Euryischia* were reared from the puparia of parasitic Diptera ; those of *Myiocnema*, from parasites of scale insects ; and those of *Eurymyiocnema*, from the parasites of mealybugs. The records indicate Australia as the centre of origin of these species. In the collection at the University of California Citrus Experiment Station there are also several undescribed species, collected in Australia, closely related to *Myiocnema* and *Eurymyiocnema*.

The three genera under consideration here, namely, *Eurymyiocnema*, *Myiocnema*, and *Euryischia*, can be separated from typical Aphelinids and Elasmids, by the characters of the front wings and the chaetotaxy of the head and thorax. The front wings are distinctive and unlike the wings of either the typical Aphelinids or the Elasmids. They are furnished with one or more erect, long, coarse bristles which arise from either the blade or the veins ; the submarginal vein is short and not broken at the junction with the marginal vein ; the marginal, postmarginal, and stigmal veins are relatively long ; a short stub of a basal vein is present in *Euryischia* and *Myiocnema*, and a vestige of such a vein is indicated in *Eurymyiocnema*. The characters mentioned above are shown best by illustrations. The wings of *Eurymyiocnema aphelinoides*, sp. n., are shown in fig. 1 ; those of *Myiocnema* have been illustrated previously by Smith and Compere (1928) ; those of several species of *Euryischia*, including the genotype *lestophoni* Riley, have been illustrated by Riley (1889), Masi (1907), and Silvestri (1914) ; and those of *Elasmus flabellatus* (Fonsc.) have been illustrated by Masi (1907).

In *Elasmus* the front wings are distinctive ; the marginal vein is exceedingly long ; the postmarginal and stigmal veins very short ; and the coarse bristles which stand out at an angle to the blade, and which characterise *Euryischia*, *Myiocnema*, and *Eurymyiocnema*, are absent.

Euryischia resembles *Elasmus* in its elasmid-like metanotum and propodeum, enlarged, compressed hind coxae, and row of coarse bristles on the dorsal margin of the hind tibiae. With these points of resemblance, the similarity between the two ends. The head, mouth parts, thorax, wings, and five-segmented tarsi of *Euryischia* are unlike the corresponding parts in *Elasmus*. In the latter genus the cutting edge of the mandibles is subdivided into five or more small teeth, the maxillary palpus is

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two-segmented, the labial palpus one-segmented, and the thorax is elongated and without parapsides; in one species at hand the axillae are absent, and the prepectus is almost divided, appearing on either side as a large triangular plate. In contrast, in *Euryischia,* as well as in *Myiocnema* and *Eurymyiocnema,* the mandibles have two

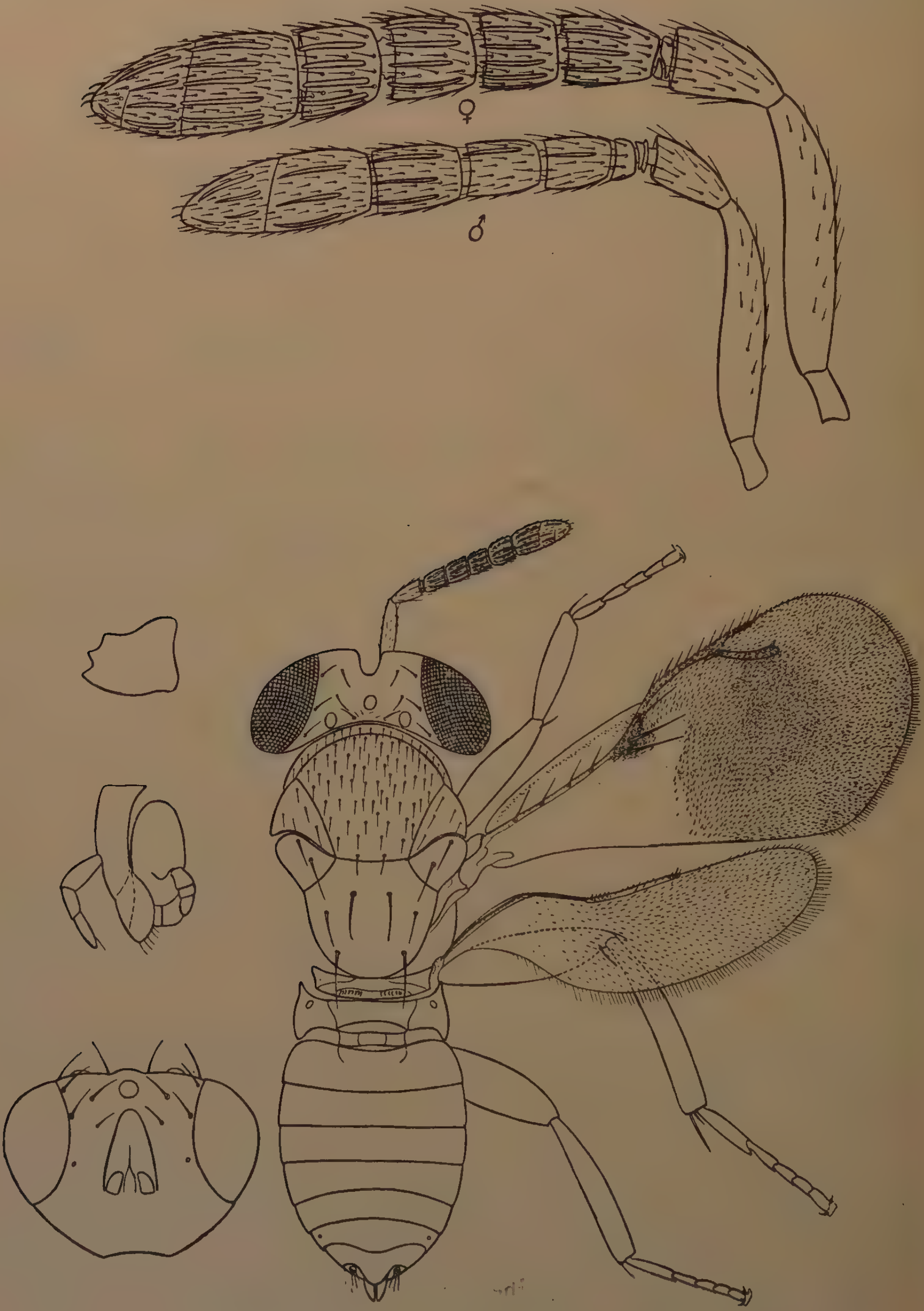

Fig. 1.—*Eurymyiocnema* (gen. n.) *aphelinoides*, sp. n.

teeth and a truncation ; the maxillary palpus is three-segmented ; the labial palpus two- or three-segmented ; the thorax is not elongated, and the parapsides and axillae are well developed ; the prepectus is not so greatly constricted across the venter as to appear divided into two parts, but is transverse, more nearly as in *Aphelinus*.

Myiocnema lacks all the characteristic features which distinguish *Elasmus*. It has been placed in the ELASMIDAE because of its relationship to *Euryischia*. *Eurymyiocnema* also lacks the distinctive characters which distinguish *Elasmus*.

Eurymyiocnema, gen. nov. (figs. 1–4).

This genus is most closely related to *Myiocnema* Ashmead. It is distinguished from *Myiocnema* by the following characters : antennae with four segments in the funicle, in addition to the ring-joints ; scutellar axillar sutures not forked so as to divide the axillae into two parts ; hind coxae not at all enlarged ; hind tibiae without coarse, stiff setae on the dorsal margin ; scrobes deeply impressed, forming an indentation, visible when the head is viewed in dorsal aspect ; basal vein vestigial, hardly indicated.

♀.—Head plainly menisciform ; about three times as wide as long, and one and one-half times as wide as deep ; face convex ; scrobes confluent and deeply indented, so that the concavity is visible from above ; antennae inserted close together and on a level with the base of the eyes ; ocelli in an obtuse triangle ; dorsum of head with coarse setae arranged in a pattern as shown in fig. 1 ; eyes bare. Antennae, exclusive of the radical and ring-joints, nine-segmented ; club three-segmented, the apical suture faintly indicated. Both maxillary and labial palpi three-segmented. Mandible rather obscurely tridentate, as shown in fig. 1.

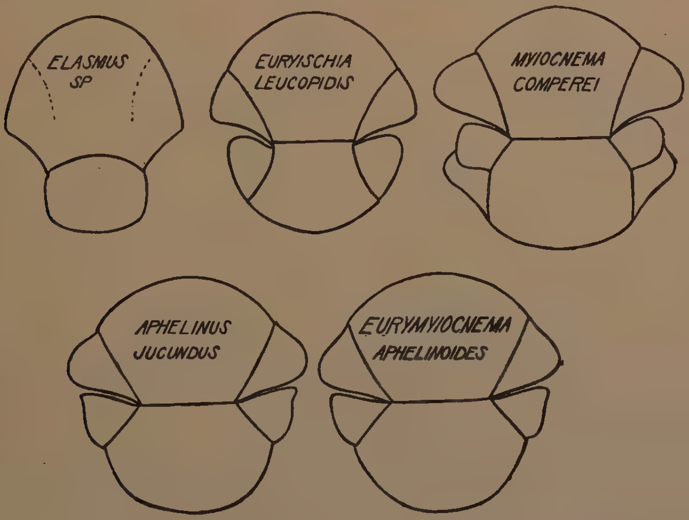

FIG. 2.—Notum of mesothorax of representative species.

Notum of mesothorax (fig. 2) about as in *Aphelinus* Dalman or *Coccophagus* Westwood, except that the postphragma (not shown in the figure) does not project into the basal part of the abdomen. Sternal and pleural components of the mesothorax indistinguishably fused ; an internal pleural ridge extends from the coxal processes diagonally to the anterior margin of the composite plate formed by the union of the sternum and pleura, as shown in fig. 3. Prepectus narrow and bandlike across the venter, slightly expanded laterally, without the armlike apodemes generally present in Aphelinids.

Metanotum, propodeum, and coxae of hind legs as in *Aphelinus* or *Coccophagus*, and as shown in fig. 4.

Outer plates of the ovipositor (gonotergites) united distally by a narrow bridge across the dorsum of abdomen, as in *Aphelinus*. Ovipositor sheaths (gonostyli) slightly longer than wide, not movable apart from the plates which bear them. Abdomen and ovipositor fundamentally similar to those of the Aphelinids.

Wings, chaetotaxy of thorax, and other details as shown in the figures.

♂.—Similar to the female, except for differences in the antennae and abdomen. First funicle joint much smaller than the following; the two ring-joints more distinctly separated than in the female, as shown in fig. 1. Fifth sternum enormously enlarged, occupying the greater part of the venter of the abdomen.

Genotype: *Eurymyiocnema aphelinoides*, sp. n.

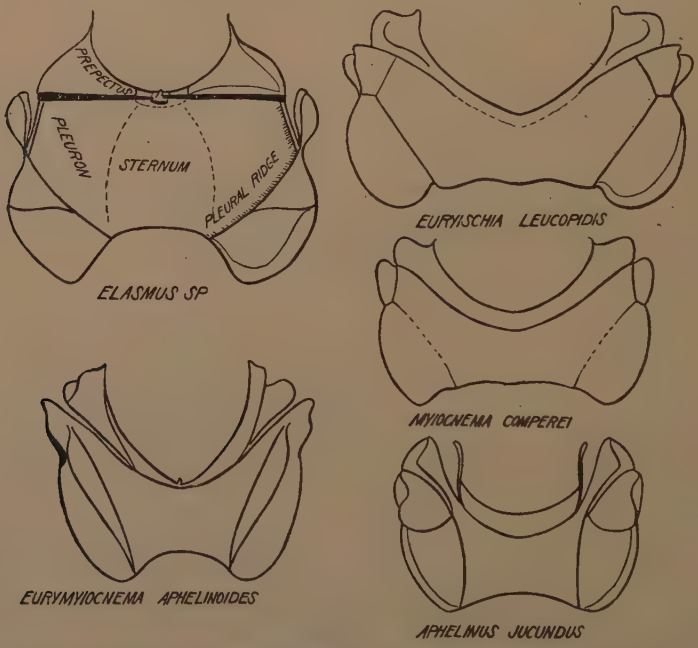

Fig. 3.—Sternal and pleural components of mesothorax of representative species.

Eurymyiocnema aphelinoides, sp. n. (figs. 1–4).

♀.—Thorax and abdomen black except for a creamy-white crossband at base of abdomen. Frontovertex and face yellowish brown; cheeks and posterior aspect of head blackish. Tibiae and tarsi yellowish brown; coxae and femora diluted with blackish.

♂.—Similar to the female except as mentioned in the generic description and as shown comparatively in fig. 1.

Length 1·6 mm.

Described from 55 specimens, holotype, allotype, and paratype. Of these 7 ♀♀ issued from mummies of *Pseudococcus* inhabited by *Leptomastix* collected by A. R. Melville at Toro, Uganda. The remainder of the material includes both ♀♀ and ♂♂

and was reared from mummies of *Pseudococcus lepelleyi* Betrem collected by R. H. Le Pelley at Buitenzorg, Java, in 1937. The holotype and allotype are from Java; paratypes from both Java and Uganda.

The first series of specimens received for identification bear the identification No. 9, and in the covering letter, dated 21st October, 1938, Le Pelley stated: "9. ex *Leptomastix* . . . emerged in Nairobi from mummies collected by A. R. Melville, in Toro, Uganda . . . Note No. 9 has been proved by rearing in Nairobi to be hyperparasitic on *Leptomastix*." The second series of specimens bear the identification No. 76, and the covering letter is dated 29th April, 1941. According to this letter, the specimens bearing No. 76 were reared from mummies of *Pseudococcus lepelleyi* collected at Buitenzorg, Java, by R. H. Le Pelley.

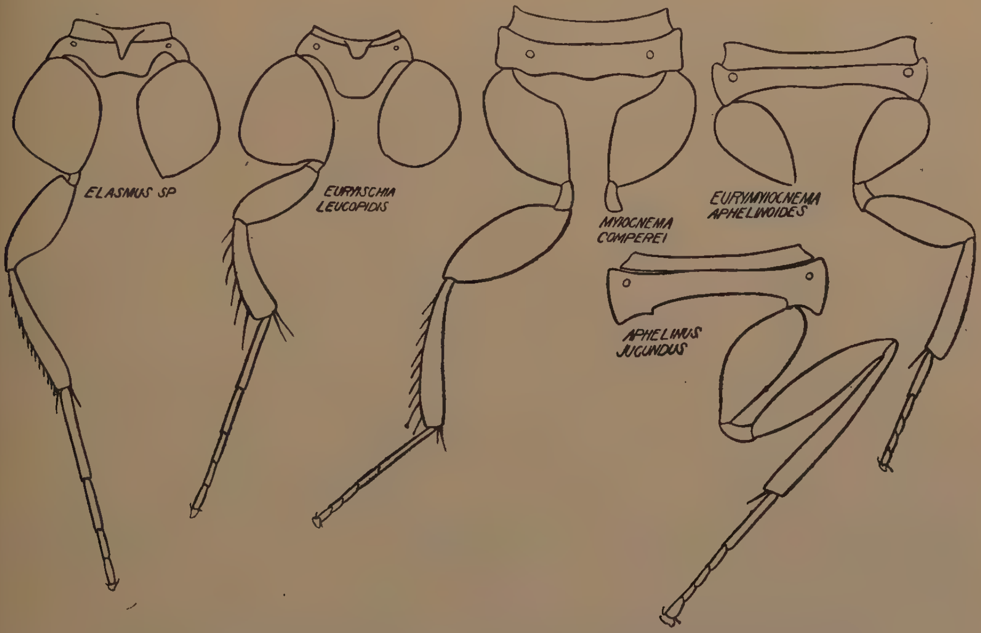

Fig. 4.—Hind legs, propodeum, and metanotum of representative species.

History of *Euryischia* Riley and *Myiocnema* Ashmead.

In an early report, C. V. Riley (1889) figured a Hymenopterous parasite under the name *Euryischia lestophoni*, and in the text stated: "This secondary parasite is a very strange form and it is illustrated at Plate IX, fig. 2. It is a new and remarkable genus of the peculiar Chalcid subfamily Elasminae, and we shall describe it in a near number of 'Insect Life' under the name *Euryischia lestophoni*, by which name it may now be called." The description which Riley proposed to publish in "Insect Life" never appeared, and the figure and brief characterisation which appeared in the 1889 report are now regarded as the original description.

Albert Koebele (1890) published an account of his observations on *Euryischia lestophoni* in Australia, and in his report reproduced Riley's figure of this insect.

W. H. Ashmead (1900) erected the genus *Myiocnema* for the Australian species *comperei*, and referred the genus to the Aphelininae. The same author (1904) classified *Euryischia* as belonging to the Elasmidae and credited the genus to Howard instead of Riley.

A. A. Girault, in a series of papers on the Chalcidoidea of Australia, expressed his opinions regarding the relationships of *Euryischia* and *Myiocnema.* In the first paper of this series, Girault (1912) wrote : " The family ELASMIDAE is a small one, consisting but of two genera. Heretofore only a single member of the family has been recorded from Australia and I have not as yet succeeded in capturing it. This species is *Euryischia lestophoni* Howard, the only representative of the second genus of the family. This genus, in my opinion, is doubtfully Elasmid since I have found an Encyrtid genus in Australia with Elasmid wings and caudal coxae and characters of this sort may have led Ashmead to ally the genus with the ELASMIDAE. However, neither the insect nor its original description is accessible to me, so that I must be content to accept Ashmead's conclusion." The " Encyrtid genus in Australia with Elasmid wings and caudal coxae," referred to above, was *Myiocnema comperei* Ashmead.

Here Girault followed Ashmead in crediting the genus *Euryischia* to Howard instead of Riley. Subsequently, the report of Koeble for 1890 came to the notice of Girault, and after that he credited the genus *Euryischia* to Koebele.

In a supplementary paper, Girault (1913a) stated : " Genus *Euryschia* Howard. The Encyrtid genus mentioned previously (I., p. 176) as bearing Elasmid coxae and wings was this genus whose position is undoubtedly correct. . . . The tarsi are 5-jointed. The antennae need careful examination, since I believe two ring-joints are present and once suspected three ; for several of the Australian species only one is recorded and, I believe, none were found in *sumneri*." Following this introductory paragraph, Girault presented descriptions of six new species, one of which, namely, *shakespearei*, was later recognised by him as a synonym of *Myiocnema comperei* Ashmead.

In a subsequent paper, Girault (1913b) defined the genus *Myiocnema* as follows : " Differs from *Encarsia* Foerster in having the hind tibiae armed with stiff black bristles." He then listed " *Myiocnema pallida* Ashmead," with the following reference : " Ashmead, 1900, Canadian Entomologist, London, Ontario, xxxii, p. 349." Girault probably did not have the original description of *Myiocnema*, published by Ashmead, but obtained this reference from a paper published by L. O. Howard (1907, p. 69) : " *Myiocnema* (n.g.) *pallida* Ashmead. Canadian Entomologist, XXXII, no. 11, Nov., 1900, p. 349. Reared by A. Craw from *Saissetia oleae* Bernard (*Lecanium oleae*), collected by Geo. Compere, Brisbane, Queensland." Apparently *pallida* was a lapsus for *comperei* by Howard.

Directly following the citation of *Myiocnema pallida* Ashmead, Girault (1913b) gives a table of genera and designates correctly, *Myiocnema comperei* Ashmead as the genotype.

Girault (1915) described the species *marmorativentris* under *Myiocnema* Ashmead. Later, he (1917) designated *marmorativentris* as the type species of the new genus *Paramyiocnema*, which he defined in the following words : " Differs from *Encarsia* in having the caudal tibiae dorsad armed with long, stiff bristles." Here it may be recalled that previously Girault (1913b) defined *Myiocnema* in the following words : " Differs from *Encarsia* Foerster in having the hind tibiae armed with very stiff black bristles."

Girault (1916) again published on *Euryischia* and *Myiocnema* as follows : " Genus *Euryischia* Koebele. *Myiocnema* Ashmead of the APHELININAE is a true synonym as I know by examining the types of the genotype of the latter. *Euryischia shakespearei* Girault. This is a synonym of *Myiocnema comperei* Ashmead, the types of which I have seen. In *antea*, *Myiocnema pallida* Ashmead (Howard) is written once for *comperei* ; the name *pallida* has no standing in nomenclature. The type number of *comperei* in the U.S.N.M. is really 5342 and not 5442 as on the specimen and as published. The species thus occurs as far south as Brisbane on the east coast and I have seen four females in the United States National Museum which were reared from *Dactylopius vastata* at Swan River, West Australia. I have specimens also from Java (van der Goot) and Koebele reared it from males of *Kermes acaciae* Maskell on Acacia at Sydney, N.S. Wales, December 21, 1899 (I have seen these specimens)."

Smith and Compere (1928) published excerpts from a letter written by A. B. Gahan, regarding the species *Myiocnema comperei* and the relationship between the genera *Myiocnema* and *Euryischia*. Gahan wrote as follows :

" In my opinion the species is not Aphelinine. It differs very decidedly in venation, especially in the long stigmal vein and the presence of a long postmarginal, as well as by having an unbroken submarginal. As you have pointed out it differs from the APHELININAE also in the legs, the hind coxae being larger and compressed, the femora also large and strongly compressed while the tarsi are long and slender. The chaetotaxy is also different from that of the Aphelinines.

" The species is certainly very closely allied to *Euryischia* although not conspecific with *E. lestophoni*. The venation of the two species is almost identical, the structure of the thorax differs but little, the chaetotaxy is similar and the general habitus of the two is very similar. *Euryischia*, however, has the hind coxae very greatly enlarged exactly as in *Elasmus*, and possesses also the peculiar triangular postscutellum of *Elasmus* and is otherwise unmistakably an Elasmid in spite of the peculiar venation. *Myiocnema* lacks the triangular postscutellum and does not have the hind coxae so large. Otherwise it differs only in minor specific characters so far as I can see. Riley's figure of *Euryischia* which you cited is inaccurate in so far as the details of the thorax you mention are concerned, as the parapsidal grooves are present and about as distinct as in *Myiocnema comperei* while the two projections on the scutellum do not exist.

" It is my opinion that *Myiocnema* is an Elasmid and whether you consider it a synonym of *Euryischia* or a good genus related to *Euryischia* is a matter of personal opinion as to the value of the characters by which the two differ. I fail to find anything more substantial to differentiate the two than the characters that I have mentioned, viz., the smaller coxae and the absence of the triangular postscutellum. The chaetae of the head, thorax and legs of *Euryischia* are longer and coarser than in *Myiocnema* but seem to be very similar in placement and number. The fore wings in both genotypes have the same short stub of a basal vein, the same long stigmal and postmarginal and comparatively short marginal. *Euryischia* has two very strong chaetae on the disk of the wing behind the submarginal vein as shown in Riley's figures but larger than he had indicated. Only one of these spines appears to be present in *M. comperei* and it is much reduced in size. *Euryischia* also lacks the patch of discal cilia basad of the stub of basal vein which you find in *Myiocnema*.

" *Elasmus* differs from both *Euryischia* and *Myiocnema* by four-jointed tarsi, a very long marginal, a very short stigmal vein, no indication of a basal, and by the absence of parapsidal grooves. These characters, on paper, seem about as substantial as those which distinguish *Euryischia* and *Myiocnema* from the Aphelinines. *Euryischia*, however, is surely Elasmid and *Myiocnema* is just as surely closely related to *Euryischia*, although the latter apparently forms a connecting link between the ELASMIDAE and the Aphelinines."

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LABORATORY STUDIES ON THE BIONOMICS OF THE RAT FLEAS, *XENOPSYLLA BRASILIENSIS*, BAKER AND *X. CHEOPIS*, ROTHS.

III. Further Factors affecting Adult Longevity.

By E. B. Edney, Ph.D.

Introduction.

The longevity of unfed adult *Xenopsylla* is known to vary with temperature and humidity, and it has also been shown (Edney, 1945) that adult longevity is also affected by psychrometric conditions during the larval and pupal stages. But a comparison of results obtained by previous workers shows that even when differences in temperature and humidity during the pre-adult and adult stages are allowed for, longevity figures vary to a much greater extent than would be expected.

Hopkins (1935) found that newly emerged, laboratory-bred fleas, kept at 20°C. in nearly saturated air, lived for an average of 21 days, and Leeson (1932, 1936) also using laboratory-bred fleas, found that adults lived 14·6 days at 18°C. and 6·5 days at 23°C., both at 90 per cent. R.H. Hopkins' conditions lie between the two sets cited from Leeson's results, so that Leeson would have obtained a figure of about 10·5 days where Hopkins obtained 21. Again, Edney obtained a figure of 28 days for longevity in the dark with unfed laboratory-bred fleas, thus agreeing more closely with Hopkins' results than with those of Leeson; but he obtained much lower figures (11·0 and 12·0 days) for other series of fleas, also laboratory-bred and kept in the same conditions.

Edney (1947) has also shown that the effect upon adult longevity of humidity during the prepupal stage is not great (relative humidities from 50 per cent. to 90 per cent. at 24°C. correspond with a range in adult longevity of about 4 days, and any relative humidity below 50 per cent. is fatal to the pre-pupal stage); so that even on the assumption that Leeson and Hopkins kept their pre-adult fleas at the extremes of the tolerable humidity range, the differences in adult longevity are still unaccounted for.

A further anomaly appears when longevity figures for wild-caught adults are compared with those for laboratory-bred fleas. The former are all low: 5·5 and 7·1 days for males and females respectively (Hopkins), and about 4 for males and 7 for females (Edney). All wild-caught fleas can be assumed to have fed at least once, and Leeson has shown that laboratory-bred adults, which are allowed to feed one or more times before being starved, live longer than those which are not fed at all. Wild-caught fleas would, therefore, be expected to live somewhat longer than unfed laboratory-bred fleas, but in fact they live for a shorter time. The figures for wild-caught fleas quoted above refer to those which have been slightly anaesthetised in removing them from their hosts, and Edney obtained somewhat larger figures for unanaesthetised, wild-caught fleas (4·2 days for males and 9·7 days for females) but the figures are still smaller than those for unfed laboratory-bred adults.

The present work was therefore undertaken with a view to finding what factors, operating either during the pre-adult or adult stages, might explain the differences found.

Material and Technique.

The material used was *Xenopsylla brasiliensis*, Baker, and *X. cheopis*, Roths., but the numbers of *X. cheopis* were not sufficient to warrant their inclusion as a separate group in the results, and they have not been used in arriving at the means. In no case, however, was there any significant difference between their behaviour and that of *brasiliensis*. Methods of breeding and of handling have already been described in Part I of these studies, and the same technique was used throughout the present work. Fleas were bred in large jars kept in a dark cupboard, the adults were fed on baby white mice more than one week old, and the larvae fed upon material in the bedding, which consisted of 100 parts of clean sand, 5 parts of bran and 1 part of dried ox-blood. The relative humidity in this bedding, measured by burying a small paper hygrometer in it, varied from 82 to 90 per cent., and the temperature in the cupboard varied from 20 to 24°C. during the course of the experiments to be described. Experimental fleas were not taken directly from the breeding jars, but were bred from eggs under controlled conditions, except where the contrary is stated.

In expressing the mean length of the various pre-adult periods the sexes have been kept separate, since it is clear from the results found that the female larval period is slightly shorter than that of the male, and the pupal period is distinctly shorter in the female than in the male. The figures for adult longevity (except when the adults were fed) are for the two sexes combined, because there appears to be no significant difference between the longevity of males and females. In 18 out of 25 series tested, the males lived slightly longer than the females, but statistical analysis of the combined probabilities, according to Fisher (1941), section 21·1, shows that the difference is not significant (P is greater than 0·1).

Experimental Results.

In Leeson's work adult fleas were obtained from culture jars, and in Hopkins' they were obtained from larvae bred singly in tubes. The pre-adult stages were therefore subjected to very different conditions (in breeding jars there are very many larvae living together, there is a mouse with its excreta, there are adult fleas and their faeces) and it was therefore decided to find whether any of those factors would account for the differences found in adult longevity.

(i) *The effect of crowding larvae.*

The first experiment was designed to ascertain whether crowding during the larval period has any effect upon adult longevity. Eggs were obtained by removing all adults from the breeding jars and leaving them overnight in a beaker, lined with black paper, in a dessicator at 80 per cent. R.H. and 24°C. The adults were returned to the breeding jar the following morning and the eggs and subsequent larvae, pupae and adults were all kept at 80 per cent R.H. and 24°C. The larvae, upon hatching, were divided into two groups. One group was divided into batches of ten each, and each batch was kept in a small test tube, 3 in. $\times \frac{1}{2}$ in., with about 1 g. of the food mixture described above. (Examination of the material left in the tube after all the larvae had pupated showed that there was plenty of dried blood still unused, so that the larvae did not go short of food.) The larvae in the other group were kept separately, each in a 3 in. $\times \frac{1}{2}$ in. test tube, with 1 g. of the food mixture. The whole procedure was repeated three times, using eggs derived from adults in different culture jars. The larvae were examined daily, and when they spun cocoons each was transferred separately to a clean tube. Each adult on emergence was put separately into a clean tube and examined daily until it died. The longevity of each flea in this and all subsequent experiments was taken from the day of emergence until the day it was found dead; thus a flea living for anything over ten days would be recorded as having lived for eleven days.

The results are shown in Table Ia, from which it is clear that crowding during the larval stages does have an effect upon adult longevity. In one experiment only, the third in Table I, was the length of the larval and pupal periods of insects kept in batches of ten measured for comparison with the lengths of those periods in insects kept separately. The results (see Table Ib) show that there is no significant effect in either sex on either the larval or cocoon periods.

TABLE Ia.

The effect upon adult longevity of crowding during the larval period.

Larvae kept together				Larvae kept separately		
Number of adults used	Mean longevity (days)	Standard deviation	P	Number of adults used	Mean longevity (days)	Standard deviation
30	16·7	4·1	$<0{\cdot}01$	22	21·0	2·1
70	15·3	5·1	$<0{\cdot}01$	36	25·8	4·3
22	18·4	4·1	$<0{\cdot}01$	20	25·0	5·4

TABLE Ib.

The effect upon length of pre-adult stages of crowding during the larval stage.

	Larval period (days)		Cocoon period (days)	
	Male	Female	Male	Female
Larvae kept together	13·2	11·7	19·3	15·3
Larvae kept separately	12·2	12·0	20·6	15·6

(ii) *The effect of the presence of adult fleas' faeces in the larval diet.*

It has been shown by Sharif (1937), Sikes (1931) and others, that blood is an essential constituent of larval food. It is obtained in the wild state from the faeces of adult fleas, which contain a certain amount of undigested blood. Since one of the differences between flea culture in jars and separate tubes is the presence of adult flea faeces in the former, it was decided to determine whether the presence of faeces during the larval stage has any effect upon adult longevity.

Eggs for this experiment were obtained as above, and when the larvae hatched they were divided into three groups. Those in the first group were placed separately in clean tubes with food. Those in the second group were placed separately in tubes with food and flea faeces. (These tubes had been prepared by keeping several adult fleas in each for the previous night. The adult fleas had defaecated and oviposited, the eggs were removed and the faeces left.) The larvae in the third group were placed separately in tubes with flea faeces and each was transferred, together with the food material, to another tube with fresh flea faeces each day during the larval period, so that the larvae in this group had a constant supply of freshly deposited flea faeces.

The results (see Table II), showed no significant difference in the length of larval or cocoon periods or in adult longevity.

TABLE II.

The effect upon length of larval and cocoon period and upon adult longevity of the presence of adult flea faeces in the larval food.

	Mean length of larval period (days)		Mean length of cocoon period (days)		Number of adults	Mean longevity of adults (days)	
	Males	Females	Males	Females			P
Faeces present at beginning of larval period	17·0	15·5	22·0	17·6	36	17·4	0·5
Fresh faeces added daily	14·8	13·8	19·5	14·8	35	18·2	0·7
No faeces	16·8	15·7	23·0	17·8	39	17·8	0·5

(iii) *The effect of contamination of the food mixture by mice.*

For this experiment three groups of larvae were used, but all the larvae in each group were kept together in a small beaker, and the cocoons were separated into clean tubes as soon as they were formed. The first group was kept with fresh, sterile, food mixture, the second with food which had been used in a breeding jar (where mice were present) for three weeks, and the third group had food mixture from the breeding jars which had subsequently been sterilised by baking at 150°C. for six hours. All stages were kept at 80 per cent. R.H. and 24°C. The results (see Table III) show no effect upon adult longevity, though the proportion of the total number of larvae which survived is very much lower in the contaminated food group than in the other two.

TABLE III.

The effect of contamination of larval food (by mice) upon development and adult longevity.

	Number of larvae	Number of adults	Mean longevity of adults (days)	Standard deviation
Fresh food (sterilised)	75	70	12·0 (P=0·6)	3·2
Contaminated food	79	18	11·5 (P=0·8)	2·4
Contaminated food subsequently re-sterilised	76	64	11·8	3·6

(iv) *The effect of age of parent upon longevity of adults of the F_1 generation.*

It was often noticeable that the first larvae to hatch after a new culture of recently emerged adults had been started appeared large and vigorous, while those hatching two or three weeks later from the same culture, if no fresh adults were added, appeared smaller and less vigorous ; and it was decided to find whether the age of the parent had any effect, not only upon size of larvae produced, but also upon longevity of the F_1 adults.

For this experiment, two series of eggs were obtained, the first from adults all less than ten days old, and the second from the same adults when they were all more than eleven days old. The larvae, upon hatching, and the subsequent pupae and adults, were kept each in a separate tube. The results are shown in Table IV, and it is clear that the age of the parents does affect the longevity of unfed F_1 adults.

TABLE IV.

The effect of age of parent upon duration of F_1 pre-adult stages and upon longevity of F_1 adults.

	Larvae							Cocoon period							Adults		
	Male				Female			Male				Female			Both Sexes		
	No. used	Duration (days)		P	No. used	Duration (days)		No. used	Duration (days)		P	No. used	Duration (days)		No. used	Duration (days)	
		Mean	S.D.			Mean	S.D.		Mean	S.D.			Mean	S.D.		Mean	S.D.
Parents less than ten days old	37	18·1	3·1	0·03	37	16·6	2·8	37	17·9	2·7	<0·01	37	14·6	2·9	74	18·0	5·9
P.		<0·01				<0·01			0·6				0·6			<0·01	
Parents more than eleven days old	45	21·6	2·8	0·03	61	20·0	3·0	45	18·2	2·9	<0·01	61	15·0	2·7	106	13·5	4·1

It also affects the length of the larval period, but not the length of the cocoon period. In the first series (from young parents) three pupae did not emerge and three larvae failed to pupate; in the second series the numbers were two and three respectively, but only those larvae which subsequently became adults are shown in the table.

Attention may here be drawn to the length of the male and female larval periods, which are significantly different (that of the female being shorter) in both series. A similar effect was found in all experiments where the length of larval period was measured; the difference is small but statistically significant.

(v) *The effect of the texture of sand in which larvae live.*

For reasons which need not be detailed here, the sand used for bedding in the culture jars had been obtained from two different sources. It was always washed thoroughly with water, but it was not sifted, and the sand from one source was of a much finer texture than that from the other. It was therefore decided to find whether this was affecting larval development and adult longevity.

Six series of about 60 larvae each were used for this experiment, and the insects in each series were kept together in a beaker at 24°C. and 80 per cent. R.H. The sand used in each series was as follows:—

(i) coarser sand;
(ii) fine sand;
(iii) fine sand after heating strongly over a bunsen flame to destroy any organic matter;
(iv) a coarse fraction of the fine sand;
(v) a fine fraction of the fine sand;
(vi) coarse sand ground very fine.

The coarse fraction used in series (iv) was obtained by washing the fine sand through a sieve with ten holes per cm. and using only what was retained. The fine grained fraction used in series (v) consisted of particles which passed through a sieve with 40 holes per cm., but the smallest particles were of colloidal size and would remain in suspension in water. No attempt was made to measure their size accurately. The same proportion of bran and dried blood was added to each kind of sand.

The results are shown in Table V, and it is clear that the texture of sand used affects both the larvae and the longevity of the adults derived from them. The duration of the larval period is not altered, but the proportion of the whole number which survive and spin cocoons is very much reduced by fine grained sand. In series (v) and (vi), when all the grains were very small, all the larvae died after a few days; in series (ii), in which the original fine sand, containing a proportion of larger particles, was used, 12 out of 56 larvae survived and pupated. The results of series (iv) and (vi) by themselves leave no doubt that it is the size of grain that matters and nothing else; in series (iv), where the coarse fraction of the fine sand was used, the larvae developed satisfactorily and the adults were correspondingly long-lived, and in series (vi), in which the larvae were kept in the coarse sand ground to a fine texture, they all died.

To find the effect of the texture of the sand on larval development larvae were weighed on the day of cocoon formation. (They can be caused to emerge without damage from their newly formed cocoons by slight agitation or pressure.) In series (ii) and (iii) only a few larvae spun cocoons on each day, and the weights are therefore only approximate, but there is no doubt that they are much lower than the corresponding weights of series (i) and (iv). These results will be discussed further below.

TABLE V.

The effect of the texture of the sand in which larvae live upon development and adult longevity.

No. of eggs which hatched	No. of larvae which pupated	Mean length of larval period (days)		Mean weight of larvae at cocoon formation (mg. × 100)	Mean length of pupal period (days)		No. of adults	Mean longevity of adults (days)	Standard deviation	Type of sand used
		M	F		M	F				
56	56	15·7	14·5	3·50	17·0	14·3	54	17·2	4·6	Coarse
56	12	17·8	15·7	1·50	17·0	13·4	11	7·3	2·7	Fine
52	10	16·0	15·0	1·40	17·0	15·6	10	6·3	2·1	Fine, ignited
46	40	15·3	14·4	3·55	19·4	15·3	38	19·0	5·1	Coarse fraction of fine sand
56	0									Fine fraction of fine sand
52	0									Coarse sand ground to fine texture

(vi) *The effect on larvae and adults of different methods of drying blood used for larval food.*

Sharif (1937) has shown that blood proteins are rendered much less useful to flea larvae if they are denatured by heat. Previous workers (including the present writer) have not usually described in detail how they dried the blood used for feeding flea larvae. Leeson used " dried " liver and Hopkins used " dried " ox blood. In view of Sharif's results an experiment was carried out to find whether different methods of preparing blood would affect, not only larval development, but also the longevity of adults derived from them.

Three series of insects were used; the larvae of the first were fed on ox blood dried at 150°C. for 24 hours, those of the second on blood which had been dried at room temperature in a vacuum over sulphuric acid, and those of the third on blood which had been vacuum-dried as in the second series but subsequently baked at 150°C. for 24 hours. Each series consisted of about 50 insects, and those in each series were kept together in a beaker with the usual proportion of bran and sand.

The results are summarised in Table VI, and they show clearly that the effect of drying the blood by heating it at 150°C. as compared with drying at room temperature is nearly to double the length of the larval period, and to reduce adult longevity from 26 to 14 days. The results of the third series, in which the blood was first dried at room temperature in a vacuum over sulphuric acid and then baked, suggest that this process does not alter the constituents of the blood to the same extent as baking fresh blood. There is no effect of these larval diets on the duration of the cocoon period, but the weight of larvae at the time of cocoon formation is much reduced by the use of baked blood.

(vii) *The effect of feeding once or several times on adult longevity.*

Although the results described above throw light on the different values for adult longevity obtained by various workers, they do not readily explain the very low figures for longevity found in wild-caught fleas; particularly in view of the fact that all wild-caught fleas must have fed at least once and that Leeson found unfed fleas to live for a shorter time than those which had fed once or several times. Before investigating the matter further it was decided to confirm Leeson's results, and experiments were carried out using newly emerged adult fleas which had been bred in the culture jars.

One day's emergence, consisting of 270 adults, was divided into three groups; those in the first group were put separately into test tubes; those in the second group were kept in sterile bedding in a large jar for 36 hours, after which a baby white mouse was added and left for one night, and the fleas which had fed were removed the following morning and kept in separate tubes. Those in the third group were kept with a baby mouse for eight days, after which they were removed and put into separate tubes. All the tubes were kept at 24°C. and 80 per cent. R.H. and were examined daily.

The whole experiment was repeated, using an adult mouse instead of a baby mouse, and the fleas in each of the series (ii) and (iii), which were fed once and for eight days respectively, were kept all together in a specimen tube during the period of feeding.

Lastly, the experiment was repeated using a rat for food. This time all the fleas were kept in separate tubes and fed separately by holding the tube upside down on the belly of the rat.

The results of this experiment are shown in Table VII, where the longevity is shown in days after emergence for the unfed fleas and in days after the last feed in the other series. They do not confirm the results obtained by Leeson, for they show that newly emerged, unfed adults live longer than adults which have fed once, and

TABLE VI.

The effect of the method of drying blood used for larval food, upon development and adult longevity.

	No. of larvae used	No. of larvae which pupated	Mean length of larval period		Mean weight of larvae at cocoon formation (mg. × 100)	No. of pupae	Mean cocoon period (days)		No. of adults	Mean longevity of adults	Standard deviation
			M	F			M	F			
Blood dried in air at 150°C. for 24 hours	64	42	30·3	27·0	0·25	42	23·6	18·0	39	13·9	3·6
P			<0·01	<0·01			>0·5	>0·5		<0·01	
Blood dried in vacuum over sulphuric acid	63	51	16·2	14·5	0·41	51	22·9	19·4	43	26·1	5·2
P			<0·01	<0·01			>0·5	0·4		<0·01	
Blood dried in vacuum over sulphuric acid and subsequently baked at 150°C. for 24 hours	64	49	21·6	19·9	0·35	49	23·8	17·8	46	18·6	5·0

the latter, with the exception of one group, live longer than those which have had the opportunity of feeding for eight days. The exception occurs in the series which were fed upon adult white mice, in which the fleas in each group were kept together, when those insects which had fed once lived for a slightly shorter time than the females of those which had fed several times. This difference is not statistically significant, and furthermore the validity of the results is doubtful, because the fleas became dirty and sticky, as a result of being kept together in one bottle. Longevity was thus curtailed and the limiting factor was probably the degree of crowding.

More importance can be attached to the results of the first and third experiments, where the insects were kept separately, large numbers were available and the differences are clear-cut.

The results do confirm Leeson's finding that there is no difference between the longevity of adult males and females if the insects are unfed or only fed once, and that there is a difference in longevity between the sexes if the insects have fed several times.

(viii) *Longevity of unfed, wild-caught adults.*

The results obtained in the last experiment provide an attractive explanation of the shortness of the survival time of adult fleas taken from wild rats as compared with that of adults bred in the laboratory. Those taken from rats have in all probability fed at least once, and according to the present results can therefore be expected to survive when starved for a comparatively short time. But if this hypothesis is correct, wild fleas caught before they have fed on a rat should be capable of surviving for a longer time than those normally caught from rats.

This hypothesis was tested by searching for pupae in rats' nests and keeping the adults which emerged, in laboratory conditions, to find their longevity. A large number of rats' nests from grass roofs of huts in the Kampala area of Uganda was collected, and as each nest was brought into the laboratory it was carefully searched for adult fleas, which were destroyed or kept for comparison as regards longevity with the newly emerged adults which it was hoped to obtain. After removing all adults the nest material was kept in a jar with a gauze cover and examined every day to see whether any adults had emerged. (The process of searching through the nest material for cocoons was found to be much too laborious when many nests had to be examined.) A few of the nests gave a fairly rich supply of adults after a few days, and as each insect emerged it was put in a tube and kept at 24°C. and 80 per cent. R.H. to find its longevity without food.

The results obtained were as follows: from three nests obtained on 21.3.46, 46 adults had emerged by 13.4.46, and their mean longevity was 11·1 days (standard deviation 4·0 days); there was no significant difference between the sexes. Now the interval between 21.3.46 and 13.4.46 is 23 days, and since the length of the pupal period under laboratory conditions is seldom more than 23 days, it is reasonable to suppose that a few of the fleas, when taken with the rats' nests, were present as larvae. The majority, however, were pupae, and probably affected only slightly, if at all, by the change from wild to laboratory conditions. It is considered, therefore, that the figure of 11·1 days obtained for this group of adult fleas is fairly close to the value which would have been obtained had the whole cocoon pre-adult been passed in the wild state.

Longevity tests on adult fleas caught from the rats' nests as they were brought in, and which had fed (the gut contained undigested blood) gave a mean figure of 6·2 days for 13 males and 6·8 days for 24 females. These figures agree reasonably well with the figures found by Hopkins (1935) and Edney (1945) for wild-caught fleas, and are significantly smaller than the mean longevity of unfed adults derived from wild-caught pupae.

TABLE VII.

The effect of feeding upon adult longevity.

Food used for adults	Unfed			P	Fed once			P	Fed for 8 days						
									Males				Females		
	No. of adults	Mean longevity (days)	Standard deviation		No. of adults	Mean longevity (days)	Standard deviation		No. of adults	Mean longevity (days)	Standard deviation	P	No. of adults	Mean longevity (days)	Standard deviation
Baby white mouse : Fleas kept in bedding during feeding period	90	24·0	6·3	<0·01	50	18·0	5·9	<0·01	13	11·0	4·5	<0·01	28	6·4	2·3
Adult white mouse : Fleas kept together in specimen tube during feeding period	46	12·4	3·4	<0·01	42	6·5	2·1	0·35	23	7·0	2·1	<0·01	27	4·0	2·1
Adult rat : Fleas kept separately in small bottles during feeding period	90	21·8	7·2	<0·01	96	15·9	4·8	<0·01	50	10·25	4·3	0·05	36	8·7	3·1

It was hoped to confirm these results by feeding adults which emerged from pupae taken with the rats' nest material, after which it was expected that they would live for a shorter time. Adults emerging on 15.iv.46 from the same nests were allowed to feed once on an adult white mouse, after which their mean longevity was found to be 14·3 days (standard deviation 3·3 days), which is significantly longer than that of the unfed adults (P is less than 0·01). This result is contrary to expectations, but its significance is very doubtful, for the adults concerned, which emerged on 15.iv.46, must certainly have spent a week or more of larval life in the nest material under laboratory conditons, and it is possible that this affected their capacity for survival more than the single feed.

Discussion.

The experiments described above were undertaken in an attempt to find the cause of certain anomalies both in the writer's previous results and in the results of other workers, regarding longevity figures for adult fleas. Certain factors have been found which operate during the larval period and profoundly affect the capacity for survival of adults, and it remains to be seen whether these factors are capable of throwing light on the particular cases where results have differed.

The difference between the results of Hopkins and of Leeson is fairly clear-cut and may be examined first. Hopkins obtained, both with *X. cheopis* and *X. brasiliensis*, consistently higher figures for survival time of unfed laboratory-bred adults (about 21 days) than those of Leeson (about 10 days). But Hopkins' larvae were reared separately in clean tubes, while Leeson's were reared in large numbers in culture jars. The foregoing experiments have shown that crowding larvae has the effect of reducing the period of adult survival, in one series from 25·8 to 15·3 days, the mean reduction of the three series being 32·7 per cent. of the higher value. A reduction of 32·7 per cent. in Hopkins' 21 days brings the figure to 14·4 days, which is still higher than Leeson's figure. But Leeson used " fine, sifted sand " as bedding in his breeding jars while Hopkins' flea larvae were kept in bran without sand, and the present experiments have shown that fine sand during larval life reduces adult longevity very considerably. So that the known differences between the breeding techniques of Hopkins and Leeson would by themselves account for differences in their results, though there were probably other differences in technique (for instance the method used for drying the blood for food is not stated) modifying the results in one way or another.

The results of Bacot and Martin (1924) also need some explanation. Working with laboratory bred fleas these authors obtained adults which when starved lived for an average of 6·3 days at 32°C. and 89 per cent. R.H. (Sat. def. 4·0 mm. Hg.), and for 2·8 days at 32°C. and 72 per cent. R.H. (Sat. def. 10·0 mm. Hg.). At 21°C. and about 47 per cent. R.H. (Sat. def. 10·0 mm. Hg.) they found adult fleas lived for 4·8 days. These figures, making allowance for conditions of temperature and humidity, are all much lower than those of Hopkins and somewhat lower than those of Leeson. Webster (1930) found unfed, laboratory bred *X. brasiliensis* to live for 5 days (females) and 7 days (males) at 27°C. and a saturation deficit of 0·3 mm. Hg., figures which are lower than Hopkins' and about the same as those of Leeson.

Bacot and Martin say little of the conditions in which their larvae were reared, except that they were bred together in breeding cages ; but they used a mixed population of fed and unfed males and females for longevity experiments and kept them in groups of ten each in small tubes, and these factors, taken together, would certainly reduce the adult longevity sufficiently to account for the difference from Hopkins' results.

The results obtained by the present writer and recorded in part I of these studies may now be re-examined. Laboratory-bred adults were found to live for as long as

28 days in one experiment (Table II) and for 12 days and less in another (Table VI). These results are understandable when it is known that the larvae in the first quoted experiment were bred singly in tubes while those in the second were bred in crowded breeding jars, that no record was then kept of the type of sand used for the larvae to live in, and that the blood used for larval food was not always baked at the same temperature for the same time. Each of these factors has been shown in the present experiments to affect adult survival.

Perhaps the most important discrepancy in results hitherto published is that between Bacot and Martin's (1924) results, where a linear relation was found between saturation deficit and adult longevity, and those of Leeson, where no such relationship was found. Johnson (1942) has pointed out that some of the discrepancies between the results of these workers are in fact due to errors in curve fitting, and that Bacot and Martin's contention that longevity is inversely proportional to saturation deficit at a constant temperature is not a valid conclusion from their experimental results. Bacot and Martin's figures are fitted best by a hyperbola-shaped curve, and they were obtained with fleas of mixed ages and states of nutrition. Johnson's analysis of Leeson's data for fed fleas shows that they also are fitted by a hyperbola-shaped curve. Leeson's data for unfed fleas and fleas which had only fed once are, however, best fitted for the most part by straight lines.

It would appear then that if the state of nutrition is allowed for, Leeson's results no longer conflict with those of Bacot and Martin as regards the relation between longevity and saturation deficiency, and it is only the position of the curves in relation to the origin on the graphs which differs. In other words, Bacot and Martin get consistently shorter survival times for a mixed population than Leeson for fed fleas; and this difference may well be accounted for by differences in conditions to which the larvae were exposed.

Some explanation of the mechanisms of the various factors found to operate in the present experiments may now be attempted, though the suggestions will often be conjectural.

Crowding larvae together certainly reduces adult longevity even in the presence of excess food, probably by inducing increased activity which in turn leads to increased metabolism and loss of water, so that the larvae do not reach the same size as, or possibly have a lower water content than, those reared singly. The adults from such crowded larvae would emerge as smaller insects (possibly with a lower water content) than those from larvae reared singly.

It is not easy to explain why contamination of the food by mice should reduce the number of larvae which pupate and later emerge successfully, without affecting adult longevity. It is worth pointing out, however, that the few larvae which do survive are probably the hardiest, and that a comparison of these insects with other adults is not valid, since they are a selection and do not represent the whole population.

It is also difficult to understand without further knowledge why the age of parent fleas should affect the duration of the larval period and the adult longevity of the F_1 generation, though similar effects are known in other animals.

As regards the effect of the texture of sand on larvae and upon subsequent adult longevity, Wigglesworth (1944 a & b, 1945), Alexander & others (1944) and Dennell (1945) have shown that fine, hard dusts have the effect of scratching or adsorbing the waxy epicuticle of some insects, thereby allowing water to evaporate from the exocuticle. It seems likely, therefore, that the fine particles of silica have the effect of breaking the relatively waterproof epicuticle of the larvae, and that they then lose water by evaporation and are unable to develop to the normal size. It is worth noting that the length of the larval period is not increased in very fine sand, but the weight of the larvae at cocoon formation is considerably below normal.

An attempt was made to find out whether larvae could be prevented from losing water as a result of living in the fine sand by keeping them in nearly saturated air, but this was unsuccessful because any relative humidity over 95 per cent. led to the development of various moulds which themselves affected the larvae adversely.

As regards the effect of feeding baked instead of vacuum-dried blood to the larvae, little more needs to be said. It is clear from the results of Sharif that the blood proteins, denatured by heat, are insufficient to support full larval growth. He obtained " lagging " larvae with denatured proteins and the same result has been found in the present experiments (30 days larval period with baked blood and 16 days with vacuum-dried blood). The underdeveloped larvae become undersized adults which do not survive as long as normal adults, although the poor longevity may not in this case be due to lack of water ; for although the adults are undersized, the proportion of water which they contain is not necessarily lower and it is more likely that their food reserves are insufficient.

With regard to the effect of feeding upon adult longevity, the present results appear to conflict with those of Leeson. His adult fleas lived for a longer time when starved if they had first had one or more meals than if they had not fed at all, whereas in the present work it was found that unfed adults lived longer than those which had been allowed to feed once or several times.

TABLE VIII.

Summary of the effect on adult longevity of various factors acting at different stages of the life history.

Author	Factor	Period of action	Effect on adult longevity
Edney (1947*)...	Crowding (rearing separately)	Larval ...	— —
,, ,, ...	Food contaminated by mice (uncontaminated)	,, ...	?O
,, ,, ...	Presence of adult flea faeces in larval food (absence)	,, ...	O
,, ,, ...	Blood for larval food baked in air (dried in vacuum at room temperature)	,, ...	— — —
,, ,, ...	Fine sand (coarse sand)	,, ...	— — —
,, (1947) ...	Relative humidity higher (lower)	Prepupal ...	O**
,, ,, ...	Relative humidity higher (lower)	Pupal ...	+
,, (1945) ...	Relative humidity higher (lower)	All preadult stages	Variable
,, ,, ...	Temperature higher (lower)	All preadult stages	— —
,, ,, ...	Light (dark)	Adult ...	—
Leeson (1932), Bacot & Martin (1924)	Temperature higher (lower)	,, ...	— —
Bacot & Martin (1924)	Relative humidity higher (lower)	,, ...	+
Leeson (1936) ...	Feeding once (starved)	,, ...	+
,, ,, ...	Feeding often (feeding once)	,, ...	+
Edney (1947*)...	Feeding once (starved)	,, ...	— —
,, ,, ...	Feeding often (feeding once)	,, ...	— —
,, ,, ...	Parents younger than 10 days (older than 11 days)	?	+ +

* indicates reference to present paper.
+ indicates an increase in adult longevity.
— indicates a decrease.
O Indicates no effect.
** Provided the R.H. is above 50 per cent. (see Edney, 1947).
The control for each factor considered is shown in brackets.

We now know that the conditions in which Leeson's fleas were reared lead to the production of short-lived adults, and it is reasonable to assume that this is due to lack of food or water reserves. Similarly, those adults reared for the present Experiment VIII were probably long-lived as a result of having good food and water reserves. The fact that a meal increases the longevity of Leeson's fleas and decreases the longevity of mine, suggests that a meal, besides adding to the reserves, alters the physiological balance of the insects in such a way as to make them less conservative of those reserves which they have. If this interpretation is correct, then Leeson's fleas lived for a very short time (6·5 days) when unfed because they had very few reserves, and the most important effect of a meal to them was to provide material which enabled them to live somewhat longer (9·5 days) when subsequently starved. On the other hand, my fleas started with ample reserves which were used economically and lasted for an average of 22 days if the insects were unfed, but were used comparatively quickly and lasted for only 10 days if the insects were fed, and the most important effect of a meal in this case was not the provision of food or water but the effect on the physiological balance which renders the fleas less conservative of those materials.

A parallel can perhaps be drawn with the physiological condition of insects in and out of diapause. Once diapause has been broken insects must feed or starve to death, and similarly, once an adult flea has fed it must do so again sooner than if it had not fed at all.

The present experiments on wild-caught fleas are unfortunately not conclusive, and little more can be said, except to remark that, in so far as unfed adults reared from pupae obtained from rats' nests lived significantly longer than adults which had fed and which were found in the same nests, the results confirm those found for laboratory-bred fleas. Some of the pupae were kept under laboratory conditions for a considerable time, and the results are, therefore, open to criticism, as are the further figures obtained from later adults bred from the nest material and fed.

The available information concerning the effect upon adult longevity of various factors acting at different stages in the life of fleas has been summarised in Table VIII. The effect is shown as —, nil or +, rather than in absolute figures, for the latter would have to be oversimplified and therefore become misleading. All the factors found in parts I and II of these studies, as well as relevant results of other workers, have also been included in the table.

Acknowledgements.

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My thanks are also due to Mr. G. H. E. Hopkins for the interest he has taken in the work and for much helpful criticism and advice.

Summary.

1. The paper describes work carried out to find the causes of certain discrepancies in the results of longevity experiments on rat-fleas.

2. The following factors were found to reduce significantly the survival period of unfed adult fleas :—

Rearing larvae together rather than in separate tubes ;
Parent fleas over 11 days old ;
Using very fine rather than coarse sand as a matrix for larval food ;
Drying the blood for larval food by baking rather than in a vacuum over sulphuric acid at room temperature.

3. The following factors were found to have no significant effect upon adult longevity :—

Contamination of larval food by mice and their droppings ;
Presence or absence of adult flea faeces in larval food.

4. The effect upon the weight of larvae and the duration of the larval period of some of the above-mentioned factors is described. The female larval period is shown to be shorter than that of the male under all conditions used.

5. It is confirmed that there is no significant difference between the longevity of male and female adults which have fed once or not at all, and that females which have fed frequently before starvation live longer than males.

6. Well developed, unfed, laboratory-bred adult fleas live longer than those which have fed once before starvation, and the latter live longer than those which have fed several times.

7. There is some evidence that unfed wild-caught fleas live longer than wild-caught fleas which have fed, but this needs confirmation.

8. All the available information regarding factors affecting adult longevity is summarised in a table.

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THE EFFECT OF AREA DOSAGE, SOLUTION CONCENTRATION AND DROP SIZE OF SPRAYED SOLUTIONS AND EMULSIONS OF DDT AGAINST MOSQUITO LARVAE.

By C. G. Johnson, D.Sc.
Ross Institute of Tropical Hygiene
and
W. H. Walton, B.Sc., F. Inst. P.
Ministry of Supply.

(Plates V–VIII.)

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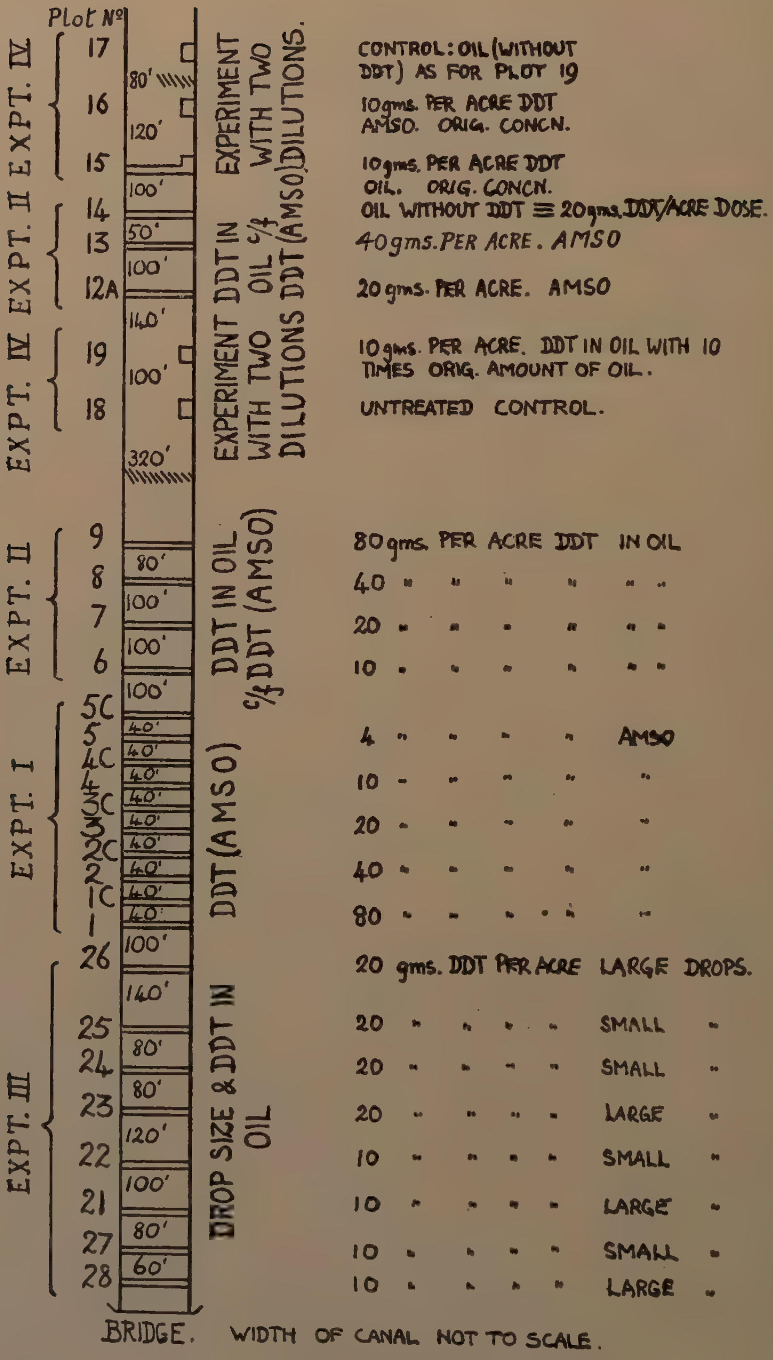
Introduction.

The work was carried out during the War and was intended to supplement the results of more practicable field trials, carried out in other parts of the world, where uncontrollable conditions and the inadvertencies associated with practical applications of insecticides would hinder precise assessment of certain variables. The object of the investigation was to assess the potential effectiveness of DDT spray against mosquito larvae living under natural conditions and to determine the characteristics of the most effective form of spray. The variables considered were (*a*) area dosage of DDT, (*b*) oil solutions compared with oil-in-water emulsions, (*c*) concentration of DDT in the spray liquid, and (*d*) drop size.

In this report the terms " area dosage " and " dose " denote the amount of DDT distributed per unit area of canal surface. The units of dosage employed throughout the paper are grams of DDT per acre. Various units have been used by other writers and the following conversion table is given to facilitate comparison of results :

10 gr./acre=2·5 mg./m^2.
=14 lb./sq.mi.
=0·176 quarts 5 per cent. solution/acre.


Plot No
EXPT. IV
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17
16
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12A
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18
9
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5C
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3C
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2C
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1C
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140'
80'
80'
120'
100'
80'
60'
EXPERIMENT WITH TWO DILUTIONS DDT IN OIL
EXPERIMENT WITH TWO OIL c/f DDT (AMSO) DILUTIONS.
DDT IN OIL c/f DDT (AMSO)
DDT (AMSO)
DROP SIZE & DDT IN OIL
CONTROL: OIL (WITHOUT DDT) AS FOR PLOT 19
10gms. PER ACRE DDT AMSO. ORIG. CONCN.
10gms. PER ACRE DDT OIL. ORIG. CONCN.
OIL WITHOUT DDT ≡ 20gms. DDT/ACRE DOSE.
40gms. PER ACRE. AMSO
20 gms. PER ACRE. AMSO
10gms. PER ACRE. DDT IN OIL WITH 10 TIMES ORIG. AMOUNT OF OIL.
UNTREATED CONTROL.
80gms. PER ACRE DDT IN OIL
40 " " " " " "
20 " " " " " "
10 " " " " " "
4 " " " " AMSO
10 " " " " "
20 " " " " "
40 " " " " "
80 " " " " "
20 gms. DDT PER ACRE LARGE DROPS.
20 " " " " SMALL "
20 " " " " SMALL "
20 " " " " LARGE "
10 " " " " SMALL "
10 " " " " LARGE "
10 " " " " SMALL "
10 " " " " LARGE "
BRIDGE.
WIDTH OF CANAL NOT TO SCALE.

Fig. 1.—Layout of experimental sections, Basingstoke canal at Odiham, 1944.

In actual aircraft trials it is difficult to apply spray of any desired characteristics to a small experimental area and so it was necessary to use a new spraying technique whereby, for experimental purposes, the spray characteristics could be accurately controlled. The choice of small experimental areas was necessary in view of the number of experiments planned and so that areas of similar uniformity in vegetation and larval population could be used.

Choice and Description of Site.

In order to make comparative experiments it was necessary to select a length of canal with a uniform type, distribution, and density of vegetation and mosquito larvae so that when divided up into experimental areas all would be as nearly identical as possible. A length of canal (approximately 1,100 yards) was chosen at Odiham (Ordnance Survey, Aldershot Command, 1 in. to mile, map ref. 175718–186713) where such conditions seemed to exist. When the canal was divided up, however, and the experimental areas were sampled, it was found that considerable variations in larval density existed from plot to plot and from day to day in the same plot, as well as slight but possibly important variations in the thickness of marginal vegetation. Thus before any differences in larval counts due to different treatments occurred there were differences due to these other factors to consider: these are discussed on pages 410–411. The experiments were made during August and September 1944. The width of the canal was about 40 ft. and each experimental area comprised a 20 ft. stretch of canal. The distance between successive areas was in no case less than 40 ft. and in most instances was considerably greater. The limits of each area were marked with numbered pegs in the bank. The disposition of the areas used in the various trials are shown diagrammatically in fig. 1. Both sides of the canal were accessible; there was a tow-path on one side, the other verge was rather marshy (see Pls. VI, fig. 2; VII, fig. 1). The canal was densely packed with vegetation. There was complete absence of water flow and no disturbance of the water surface due to human agency, *e.g.*, barges.

The central channel (sampling position 4 and 5, see fig. 3) was uniformly overgrown with *Potomogeton natans* so dense that there appeared to be more area of leaf than of open water surface (Pls. VI, fig. 2; VII, figs. 1 and 2). In places there were large masses of floating algae (a very coarse species of *Spirogyra*: Pl. VIII, fig. 1). Patches of open water in the sampling area free from either *Potomogeton* or *Spirogyra* were rare and when present were only a few inches or at the most a foot or two in extent (Pl. VIII, fig. 1).

This floating vegetation was continuous across the canal between the outer edges of the reed margins on opposite banks. At the outer edges of the reed margin there was sometimes a slight thinning out of *Potomogeton* (position 3). The reed margin varied in width from bank to outer edge from 1 to 5 ft. with an average width of about 4 ft. The dominant species here was *Sparganium ramosum* (Bur-reed). Sub-dominants were *Typha latifolia* ("Bull-rush"), *Juncus* sp., *Cyperus* and *Carex* spp. and, in places where the reeds were less dense, *Menyanthes trifoliata* (Buckbean) tended to become dominant.

The height of the vegetation of the reed margin varied between 1 and 4 ft. Except in a few places, mentioned specifically in the text, the extreme margin (position 1) where water and bank met, was completely obscured from above by densely packed reeds which had to be lifted or parted by hand to introduce the ladle when sampling.

The water surface among the standing reeds (position 2) was usually clear and free from floating vegetation, but it was much obscured from above by the standing rushes (Pl. VIII, fig. 2).

The species of mosquito were *Anopheles maculipennis*, Meigen, var. *messeae*, Flni., and some *Anopheles claviger*, Meigen.

Spraying Technique.

Spray requirements.—For the purpose of these experiments, it was necessary to devise a method of spraying known concentrations of liquid uniformly over the whole of each experimental area. Owing to the small size of the areas (approx. 800 sq. ft.) this necessitated the development of an apparatus capable of distributing a minimal dose of about 4 cc. uniformly over this area. Since it was required to investigate the effects of spray concentration and of drop size for solutions and emulsions of widely different concentrations, the volume of liquid and drop size delivered by the spray had to be independently variable over a wide range. The spray concentration required to be varied from 200 ml./acre corresponding to 10 g. DDT/acre from 5 per cent. solution, to 16 l./acre, corresponding to 80 g. DDT/acre from 0·5 per cent. emulsion, a range of 1 : 80. The smaller drops used were required to simulate aircraft spray, mean dia. c. 0·4 mm. and the larger drops to be of the order of 1 mm. in diameter.

Description of Apparatus.—The apparatus developed for the trial is illustrated in Plate V, fig. 1. It consisted of a rotating disc atomiser in which the liquid was fed from a syringe on to a motor-driven spinning disc and was thrown off from the periphery in the form of a fine spray by centrifugal force. This type of spray was chosen because the drop size was uniform, independent of the rate of liquid flow over a wide range, and was readily varied by altering the speed of the disc. The apparatus was carried on a cord stretched across the canal (Pl. V, fig. 2). The liquid delivery was determined by the movement of the syringe piston, which was carried on a screwed rod forced down by a rotating nut driven by a gear and pulley system. The carrying line passed round this pulley and the liquid was, therefore, delivered at a constant amount per unit distance travelled, independently of the speed at which the apparatus was towed across the canal. The flex carrying current to the motor was used for pulling the spray in one direction, and a separate cord was provided for pulling it back across the canal. The direction in which the cord passed round the pulley could easily be reversed for operation of the spray in either direction of travel. The apparatus was provided with interchangeable gears and pulleys giving a range of piston speeds in the ratio 1 : 2 : 4 : 8. Interchangeable syringes having cross sections in the ratio of 10 : 1 were also provided giving a total range of liquid deliveries in the ratio 1 : 2 : 4 : 8 : 10 : 20 : 40 : 80. The spray covered a band of width of from 5 ft. 3 in. to 6 ft. 8 in. according to the type of the disc used, so that four traverses of the canal were necessary to cover an experimental area 20 ft. wide.

In the first experiments, a plain disc, concave side upwards, as shown in Plate V, fig. 1, was used, but it was found that this gave a somewhat higher concentration of drops at the edges of the sprayed band than in the middle (Pl. VI, fig. 1, A) an effect which was accentuated by any slight overlap of adjacent bands. A new disc with circumferential slots allowing drops to be thrown off at two different radii was next used and gave a satisfactory distribution for "small" drops as shown in Plate VI, fig. 1, B, but was unsatisfactory at larger sizes. A "disc" with a spiral edge expanding from 0·5 to 1·75 in. radius was finally employed and gave a satisfactory distribution for both small and large drops (Pl. VI, fig. 1, C, D). It will be seen that this disc gave a falling off in concentration at the edges of the band so that a slight overlap of adjacent bands was necessary in order to secure a uniform contamination. It also made the spacing of the bands much less critical.

The dimensions of the syringes, gears and pulleys were designed so that in various combinations concentrations of 4, 10, 20, 40 or 80 g. DDT/acre could be delivered, using either 5 per cent. solutions of DDT in oil or 0·5 per cent. emulsions. In practice these dosages could not be obtained exactly with solutions of these concentrations, mainly owing to variations in the width of the sprayed belt according to the type of disc employed. The concentrations of the solutions were therefore adjusted as necessary to give the correct dosage of DDT per unit area. With the plain disc giving a 6 ft. 8 in. belt the solution concentrations required were 1·08 times the nominal values given above. For the 5 ft. 3 in. belt given by the spiral " disc", the concentrations were reduced to 0·86 of these values.

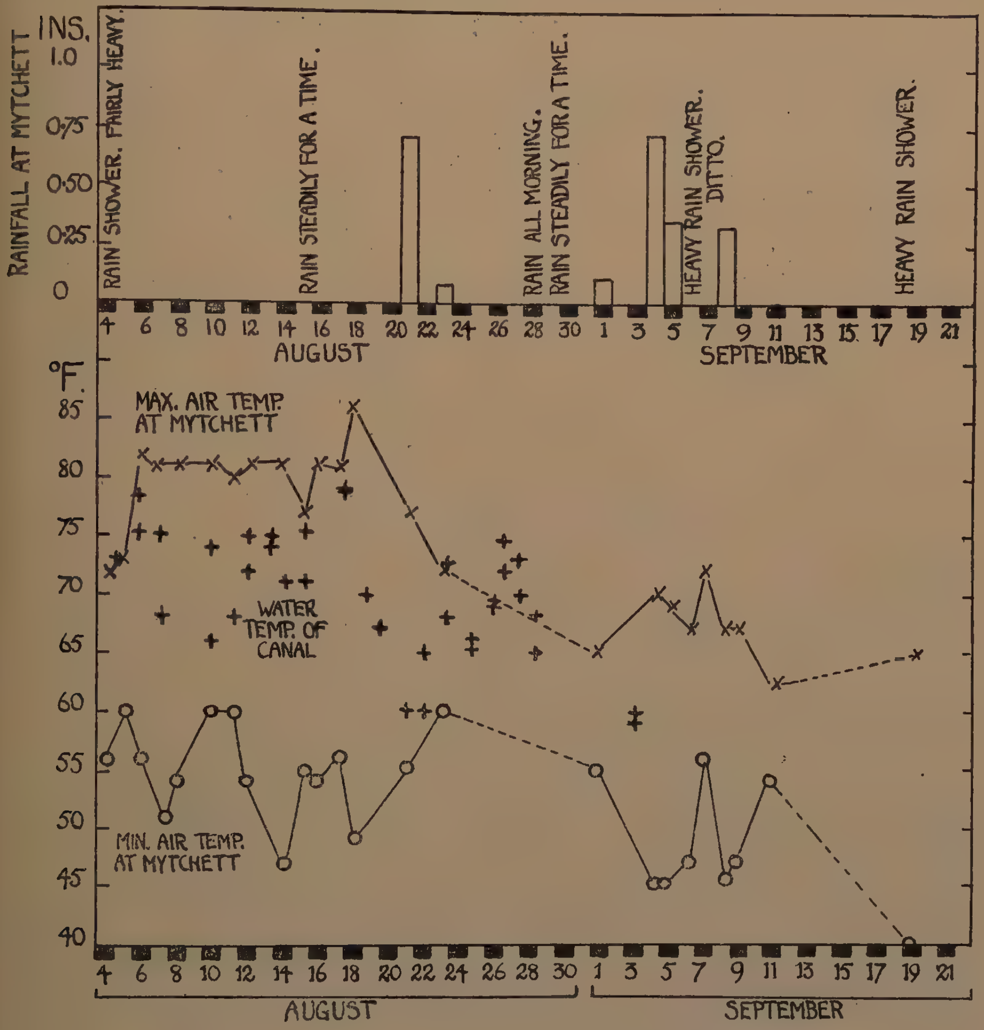

Fig. 2.—Diagram of rainfall and temperature during the experiments.

Spraying procedure.—Four men were required to operate the spray, two on each side of the canal. One man on each side supported a pole, 7 ft. high, to which the carrying line was attached. The tension of the line was adjusted to keep the spray at the correct height above the water surface. The other two men were responsible for towing the apparatus across the canal, changing the direction of the line round the pulley according to the direction of travel, and for refilling the syringe with solution when necessary. The 12 volt batteries and other electrical equipment for driving the motor and regulating its speed were situated on the tow-path side of the canal. The apparatus was moved along a measured distance equal to the band-width sprayed after each traverse of the area. It was observed that the fine spray in particular was carried an appreciable distance downwind before reaching the canal surface and so an extra band was sprayed beyond the upwind edge of each experimental area. The apparatus was also kept at the minimum height, 3–4 ft., above the water necessary to ensure a full spread of the spray. During some of the trials the wind was rather gusty. Then the spraying was interrupted as far as possible during the gusts and carried out only during the periods of comparative calm.

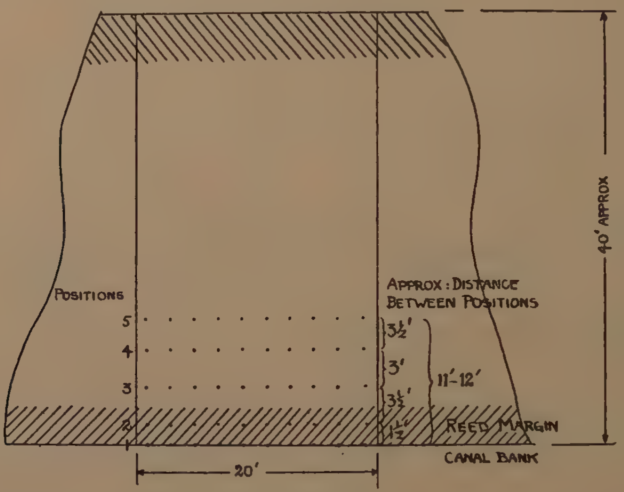

Fig. 3.—General arrangement of experimental area for sampling. Each dip is represented by a dot. Spraying was carried out over the whole width of the canal but samples were taken only from an area on one side.

Spray assessment.—The amount of solution delivered from the syringe per unit area of the experimental areas was known with accuracy, but blank runs over a sampling layout were made on a number of occasions in order to estimate the wind loss, uniformity of deposition, and drop size. The layout consisted of a row of eight floats equally distributed across the width of an area along a line parallel to the bank of the canal and about 10 ft. out between positions 3 and 4 (fig. 3). Each float carried a 9 in. × 10 in. sheet of filter paper (Whatman No. 1) on which the spray was received. For the purpose of the estimation, dyed oil or emulsion containing no DDT was sprayed. The number of drops on each paper was counted and the dye was then extracted and estimated colorimetrically. From these data the drop size and concentration of spray per unit area were calculated. The amount of spray intercepted by the reeds in the marginal areas of the canal was not determined.

Technique for sampling Larvae.

The ideal technique for estimating a population before and after spraying should be so arranged that one area is sampled several times each day. Then the fluctuations in numbers of larvae from plot to plot, day to day, and the " chance " variations in numbers counted due to random dipping, can be measured. Differences due to DDT treatments can thus be distinguished from differences due to the other factors. Unfortunately it was impossible to sample each area more than once a day, time and personnel being the limiting factors and for this reason the " sampling error " was not measured. In view of this it is idle to ask if a difference between two areas after treatment is statistically significant. For such a difference may be due, not to the treatments, but to one of the other factors mentioned above. No tests for significance have therefore been made in the following results. Where differences do exist, they are surprisingly consistent and in accordance with expectation, or else are so slight that similarities rather than differences are outstanding.

The larval and pupal population in each experimental area was estimated by taking samples with a ladle fixed to a long handle. The total number of larvae and pupae for each dip and at the same time the numbers in each of the different developmental stages (1st, 2nd, 3rd and 4th instar and pupa) were recorded. The actual dipping was standardised and the ladle always put into the water in the same way. All larvae and pupae after counting were returned to the spot from which they had been taken.

Each experimental area was sampled at five different " positions " with respect to the bank, each position corresponding to the distribution of vegetation (fig. 3). Positions 4 and 5 were in the " open " canal, position 3 was at the outer edge of the marginal reeds, position 2 was among the marginal reeds and position 1 was at the very edge, among the reeds, as close to the bank as possible.

Ten dips were taken from each of the five positions at approximate intervals of 2 ft. (making a total of 50 dips for each area) on each day for sampling. In most experiments each area was thus sampled on each of three successive days immediately before spraying with DDT (days 1, 2 and 3) : spraying was carried out on day 4, and sampling was again made on the following three successive days (5, 6 and 7) and then on single days at approximately weekly intervals.

Experiments.

The experiment was carried out in four parts, priority being given to the more important items, as it was not known at the outset if the whole programme could be completed before the end of the mosquito season. Priorities were allocated as given below. It proved possible to carry out all the tests.

First Experiment.—Determination of the minimum lethal dose with DDT in AMSO, 0·5 per cent. (approx.) emulsion, droplets circa 0·4 mm. diameter.

Second Experiment.—Comparison of DDT in oil (5 per cent. solution) and in AMSO (0·5 per cent. emulsion), droplets as in 1st Experiment.

Third Experiment.—Effect of droplet size with DDT in oil (5 per cent. solution) at delivered dosages of 10 and 20 g. DDT per acre.

Fourth Experiment.—Effect of dilution. Equal dosages of DDT (10 g./acre) delivered in solutions of various dilution. Effect of oil without DDT.

Compositions of Spray Mixtures.

AMSO Emulsion* was an oil-in-water emulsion, the oil phase of which was substantially a 5 per cent. solution of pure DDT (active isomer only) in a heavy lubricating oil.

The spray mixtures referred to in the text had the following compositions :—

(i) *AMSO Emulsion.*—The emulsion was prepared by adding distilled water to a self-emulsifying oil solution of DDT (AMSO-1) containing :—

30 g. pure DDT : cryst. m.p. 108°C.
400 ml. pool 3 oil
200 ml. Amoa A5X

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*Anti-Malaria Soluble Oil. Code name given to the self-emulsifying oil solution of DDT used in preparing the emulsion.

The emulsifying agent (Amoa A5X) was a proprietary mixture, consisting substantially of sodium oleate and cresol, which was miscible with the heavy oil, and brought about the immediate emulsification of the latter when the mixture was stirred into cold water.

The standard 0·5 per cent. AMSO emulsion was made by diluting 1 volume of AMSO-1 with 9 volumes of distilled water. For the experiment, the dilution with water was varied slightly according to the band width sprayed (see p. 408) in order to give the desired delivery of DDT per unit area. In the 1st experiment where the spray-band width was 6 ft. 8 in., the emulsion concentration was 0·54 per cent. DDT. In the others, the spray-band width was reduced to 5 ft. 3 in., and so the concentration was reduced to 0·43 per cent. in order to maintain the same area, dosage of DDT.

Distilled water was used in the preparation of these AMSO emulsions in order to secure high stability in the sprayed mixture and to avoid any possibility of bulk lack of homogeneity due to curding.

In this way it was possible to disperse on a water surface fine droplets of DDT in oil (of diameter of the order of 10μ). Small scale laboratory experiments showed that the dispersion of an emulsion in this manner gave, more rapidly, a much more even covering of a water surface with a DDT preparation than could be achieved by the direct use of a solution of DDT in oil (as below). It was felt that this use of an emulsion might be of particular advantage in dealing with water surfaces broken up by foliage which are the normal breeding places of mosquito larvae.

A subsequent mathematical investigation, however, indicated that there was little to be gained in spraying an aqueous emulsion from an appreciable height above ground, since evaporation of the water phase occurred in the falling drops.

(ii) *Emulsion of Oil Yellow.*—Used for spray assessment in 1st experiment. Composition similar to 0·5 per cent. AMSO emulsion, but containing 0·23 per cent. Oil Yellow dye dissolved in the oil phase ; no DDT.

(iii) *Oil Solution of DDT.*—Since it is difficult to dissolve technical DDT directly in heavy oil without heat or mechanical stirring, the oil solution of DDT was made by diluting a 20 per cent. concentrate of pure DDT (active isomer only) in solvent naphtha with a heavy lubricating oil to give finally a 5 per cent. solution of DDT in oil. This represented, after spraying, a saturated solution of DDT in the heavy oil, since solvent naphtha was found to evaporate rapidly when spread in a thin film on a water surface. To ensure that the heavy oil would spread on dirty stagnant water ½ per cent. of oleic acid was added to it. Thus the standard 5 per cent. solution contained :—

10 g. pure DDT : cryst. m.p. 108°C.
50 ml. heavy solvent naphtha
150 ml. HD50 lubricating oil
1 ml. oleic acid.

It is inadvisable to spray naphtha concentrate alone. It evaporates on water, and the crystals of DDT which then separate soon sink, since their density is so much greater than that of water. The density of the 5 per cent. solution of DDT in heavy oil is less than unity, and consequently the latter persistently contaminates the water surface. The concentration of DDT relative to the other constituents was adjusted slightly, as for the AMSO emulsion, according to the band width sprayed.

(iv) *Dyed Oil Solution.* Used for spray assessment in 3rd experiment. Composition as (iii) but containing 1·35 per cent. Oil Yellow dye ; no DDT.

Characteristics of the Sprays delivered.

Delivered dosages.—By a suitable choice of gears and pulleys (see p. 408), it was possible to deliver from the spray apparatus the exact area dosage of DDT required in each test. Some spray was blown away by the wind and lost, however, particularly the finer droplets, in spite of the precautions described on p. 409. There was therefore some uncertainty in the exact concentration of DDT reaching the water surface in any given experimental area. Some indication of the error was obtained by assessment trials in which dyed solutions were sprayed. The results of these experiments are given below. *For the purpose of simplifying the discussion of the entomological results throughout this Report, the "delivered dosages" of DDT will be quoted,* i.e. *the area dosage of DDT delivered from the spray. It should be borne in mind that the actual dosage on the water surface may have been about 30 per cent. less than the quoted value in the case of the fine spray ; the error was probably negligible for the coarse spray* (*p.* 414).

An error was made in making up the oil solution of DDT used in the 2nd experiment. The concentration of the solution was 25 per cent. too high (5·4 instead of 4·3 per cent.), and so the dosage actually delivered was 25 per cent. greater than the delivered dose indicated in the diagrams and text of this report. The delivered dosage of DDT in AMSO in these tests was as stated.

Drop size.—The drop-sizes of the sprays used in the various tests are given below. The assessments were made with dyed solutions sprayed over a float layout (p. 410). The number of drops and total amount of dye falling per unit area were determined as described on p. 410 and hence the drop size was calculated.

Experiments	Drop size (dia.)	Remarks
1st	0·42 mm. (" Small drops ") ...	By assessment of dyed emulsion. Plain spray disc.
2nd	0·42 mm. (" Small drops ") ...	No direct assessment. Disc speed similar to previous tests. Slotted disc.
3rd	0·41 mm. (" Small drops ") ... 0·95 mm. (" Large drops ") ...	By assessment of dyed oil solution. Spiral disc.
4th	0·95 mm. (" Large drops ") ...	No assessment. Spraying conditions as for large drops in 3rd tests.

In all cases the drops were predominantly of uniform size, as shown by the stain diameters given in the tables below. The sprayer always produced a quantity of very small drops of uniform size in addition to the main spray output of larger uniform drops. These smaller drops were the satellites formed when the parent drops detached themselves from the spinning disc. They constituted only a negligible proportion of the total mass of the spray.

Wind loss and uniformity of deposition.—In the assessment trial of the 1st experiment, three bands, 6 ft. 8 in. wide, were sprayed over the experimental area only and spraying was not interrupted during the frequent gusts of wind. Examination of the layout showed that there was a low concentration of spray over the upwind half of the area. Therefore, in subsequent runs with DDT spray, an extra band was sprayed outside the upwind edge of the area in order to allow for the spray being blown down-wind and spraying was stopped when the wind velocity increased. The assessment results with spray containing no DDT, therefore, did not accurately represent the area dosage of the DDT sprays. The average value obtained over the whole width of the plot was 38·6 per cent. of the delivered dose. When only the down-wind half of the layout is considered the spray recovery comes out at 60 per cent. of the delivered dose. The actual value for the DDT spray was, therefore, probably rather greater than 60 per cent.

Detailed assessment results for the 3rd experiment are given below. Here the dye solution was sprayed under similar conditions to the DDT spray on the experimental area, and the results therefore give a more reliable indication of the wind loss and uniformity of deposition than in the 1st experiment. It will be seen that with large drops there was no loss of spray due to wind within the limits of experimental error. About 30 per cent. of the small drops were lost.

(i) *Large drops.*

Envelope No.	No. of stains on envelope	Dia. of stains	mg. dye per m^2
A1	9 3	6 mm. 10 „	1·48
A2	16	6·5 „	1·81
A3	10	7 „	0·94
A4	11 16	7 „ 2 „	1·15
A5	28	7 „	2·96
A6	5	7 „	0·63
A7	25	7 „	2·43
A8	18	7 „	1·81

Delivered dosage equivalent to 20 g. DDT/acre in 4·3 per cent. solution.

Average droplet concentration	273 per m^2
Average dye concentration	1·65 mg./m^2
Drop diameter	0·95 mm.
Recovery. Per cent. of delivered dose	106 per cent.
Wind loss	nil
Uniformity of deposition on envelopes. (average deviation from mean value)	36 per cent.
Mean distance apart of drops	6·1 cm.

(ii) *Small drops.*

Envelope No.	No. of stains	Av. size of stains	mg. dye per m^2
B1	48	3 mm.	0·42
B2	74 110	3 mm. 1 mm.	0·63
B3	159	3 mm.	1·36
B4	67	3 mm.	0·59
B5	181	3 mm.	1·57
B6	128	3 mm.	1·30
B7	120	3 mm.	1·22
B8	167 202	3 mm. 1 mm.	1·48

Delivered dosage equivalent to 20 g. DDT/acre in 4·3 per cent. solution

Average droplet concentration	2,130 per m^2
Average dye concentration	1·07 mg./m^2
Droplet diameter	0·415 mm.
Recovery per cent. of delivered dose	69 per cent.
Wind loss	31 per cent.
Uniformity of deposition on envelopes (average deviation from mean value)	37 per cent.
Mean distance apart of drops	2·2 cm.

It was noticed that the oil in particular, and also the AMSO, even in the lowest dosages, spread right up to the bank of the canal, *i.e.* to position 1. Even the smallest corners, much obscured by reed from above, were covered with a film within 24 hours which intimately penetrated the many irregularities where the bank shelved into the water (see p. 407).

Entomological Results.

FIRST EXPERIMENT.

Determination of the minimum lethal dose with DDT in AMSO emulsion. Small drops.

AMSO emulsion concentration ...	0·54 per cent. DDT.
Delivered dosages	80, 40, 20, 10 and 4 g. DDT/acre.
Drop size	0·42 mm. diameter.

Ten experimental areas were chosen each with a space of 40 ft. between it and the next area. Alternate areas were sprayed with DDT in AMSO delivered at the rates of 80, 40, 20, 10 and 4 g. of DDT per acre. The remaining areas (with one exception) were left unsprayed, as controls. The exception was the area used for drop assessment next to that sprayed at the 80 g. rate. This area was sprayed with dyed AMSO *without DDT* at the rate equivalent to 40 g. DDT per acre. The layout of the experiments is shown in fig. 1. The total larval and pupal numbers for all dipping positions—*i.e.* from bank to central region of the canal—are shown in fig. 4. On the three successive daily counts before spraying the larval numbers varied between 47 and 93 per 50 dips. The areas were sprayed on day 4 and counts on the following days (5, 6 and 7) showed that the population had dropped in order of dosage—*i.e.* the heaviest dose, 80 g. per acre, gave the greatest kill, the smallest dose delivered, 4 g. per acre, gave the least kill (see also fig. 5).

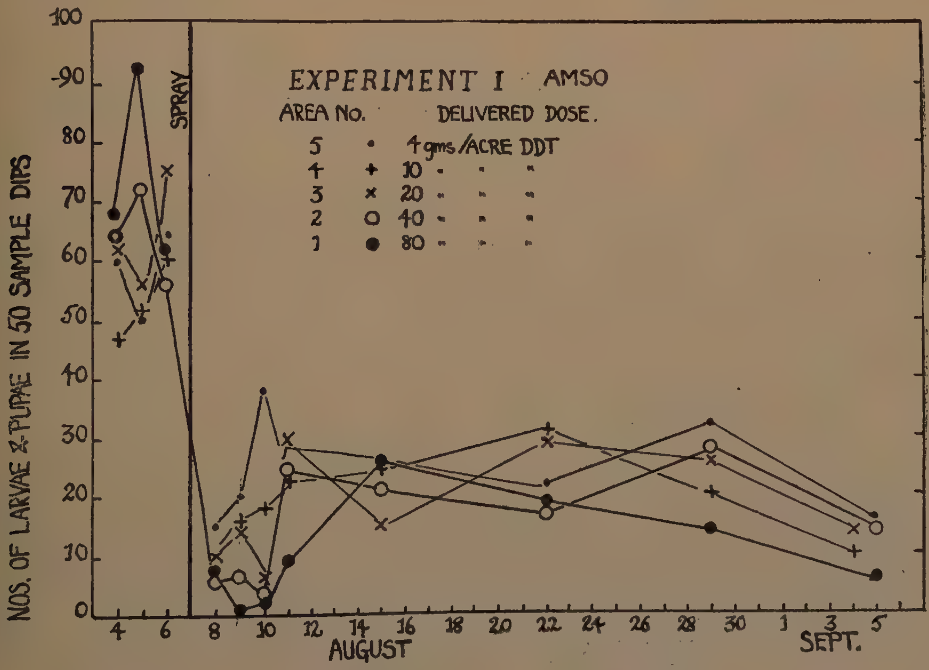

FIG. 4.—Experiment No. 1. DDT in AMSO.

The general situation is seen more clearly if the figures are put on a proportional basis. If the daily average for the three successive counts before spraying be taken as 100 per cent. and each subsequent daily count expressed as a percentage of this, we get the graphs shown in fig. 5. The degree of recovery is of the same order in each area, but the lower dosages recover to a slightly higher population level than the heavier ones because the initial kill is not so great.

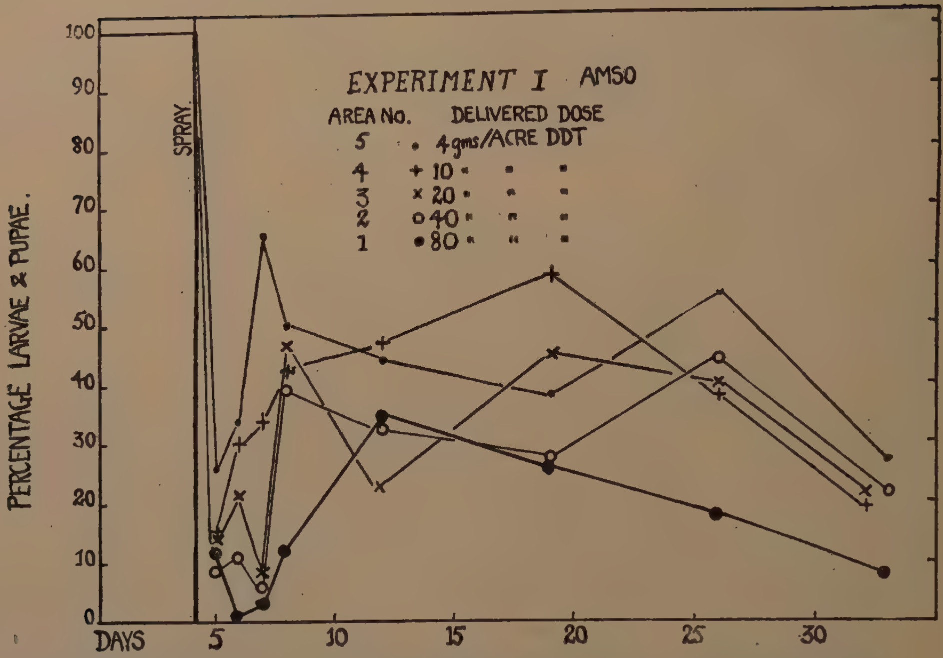

FIG. 5.—Experiment No. 1. DDT in AMSO. The numbers of larvae and pupae have been expressed as a percentage of the daily average for the three successive days before spraying.

After the 3rd day from spraying, the larval population showed an increase in all the areas, the recovery starting within 48 hours of spraying with the two lowest dosages. This recovery, due no doubt to the hatching of eggs not killed by the spray, does not continue beyond the 4th day after spraying except at 80 g. per acre where the peak of recovery is reached 8 days after spraying. The results show quite clearly that initial kill and speed of recovery are correlated with dosage. This recovery is, however, incomplete and the population never regains its original state but continues at a much reduced level until it declines at the end of the season.

This stabilisation of the population at a low level may be due to several factors and not at all to the residual effect of DDT. For example :—

(i) The reproductive potential of the population is probably insufficient to allow for complete recovery at that time of year (August–September). If the recovery seen in fig. 4 is due to the hatching of eggs already laid at a time of spraying, then subsequent egg production must have been low, otherwise a greater increase would have occurred. This is also suggested by the behaviour of the population in the control areas (see below), whose general decline after August 16th is associated with a drop in temperature (*cf.* figs. 6 and 2).

(ii) This low egg production may have been partly seasonal and partly due to some adults being killed off either by the spray while they were resting in the reed margin or due to a kill of adults when they visited the area to oviposit.

In the case of the untreated control areas (fig. 6), areas 4c and 5c, those next to 10 and 4 g. per acre areas respectively (fig. 1) show a fairly high population level over a period beyond the spraying date. Area 5c was at the end of the series and could have been influenced only by the treated area to one side of it (and this at only 4 g. per acre). It is highly unlikely that fluctuations in 4c and 5c are due to DDT spreading from neighbouring areas and all variation is almost certainly due to natural causes. Both areas 4c and 5c show a decline in larval numbers corresponding to the stabilisation level in the treated area and to a drop in temperature (fig. 2) and this supports the view that the stabilisation was due rather to natural causes than to DDT.

All the other control areas, 1c to 3c, show a decline about five days after the neighbouring areas were sprayed, and it seems almost certain that these control areas were affected by the DDT deposited 40 ft. on each side of them. But in every control area there is a slight upward trend in the population which is reflected in the recovery of the treated areas which took place at about the same time. There was evidently a natural tendency for the larval population of the canal to increase just then.

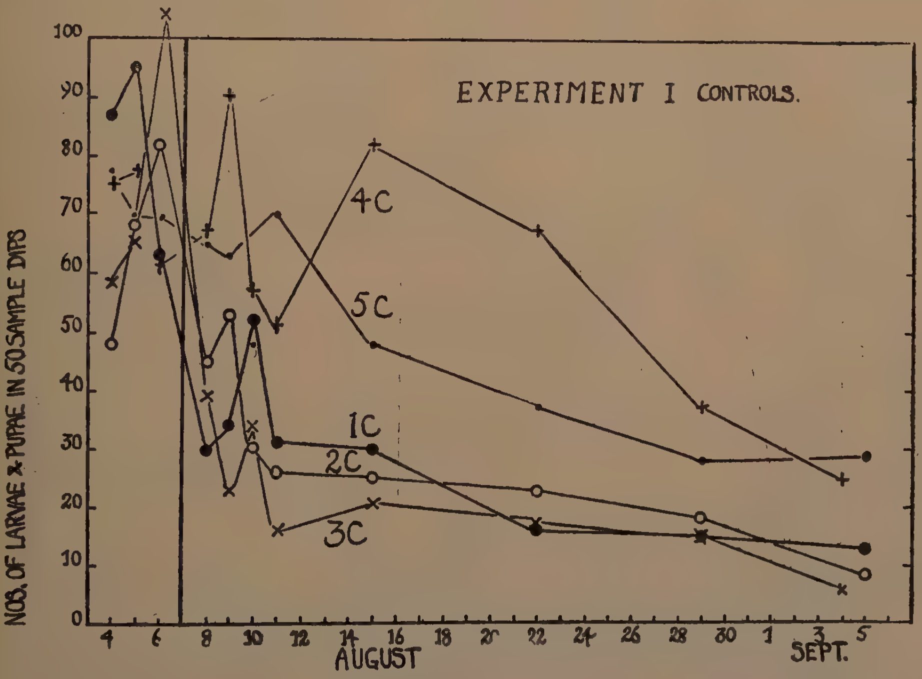

FIG. 6.—Experiment No. 1. Larval and pupal population in untreated control areas.

Thus the evidence for prolonged residual effect of DDT as a larvicide is inconclusive. The dosage for initial kills is best seen if the average daily count for the two days immediately after spraying is expressed as a percentage of the daily average for the three days before spraying (it is better not to include the 3rd day after spraying since by then recovery has started at the lower dosages). The results are shown in fig. 7 A, and are discussed below.

SECOND EXPERIMENT.

Comparison of DDT in oil and in AMSO. Small drops.

AMSO concentration ...	0·43 per cent. DDT.
Oil solution concentration	5·4 per cent. DDT.
Delivered dosages AMSO...	20 and 40 g. DDT/acre.
„ „ Oil ...	10, 20, 40, and 80 g. DDT/acre (plus 25 per cent. see p. 413).
Drop size	0·42 mm. diameter.

Seven areas were chosen: Nos. 6–9, to be treated with DDT in oil. They were arranged with 100 ft. of canal between each area except between 8 and 9 where only 80 ft. could be allowed if vegetation was to be kept fairly uniform.

The areas to be treated with AMSO and the control area (12A, 13 and 14) were 100 and 60 ft. apart and there were 320 ft. between area 9 and 12A. The general layout is shown in fig. 1.

Thus for purposes of comparison we have not only oil and AMSO treated areas in the 2nd experiment but also AMSO areas from the 1st experiment.

	Area No.		*Delivered dose of DDT in g./acre.*			
Expt. II	6–9	Oil ...	10	20	40	80
	12A and 13	AMSO ...		20	40	
	14	Control without DDT but oil≡20 g./acre area.				
I	1–4	AMSO ...	10	20	40	80

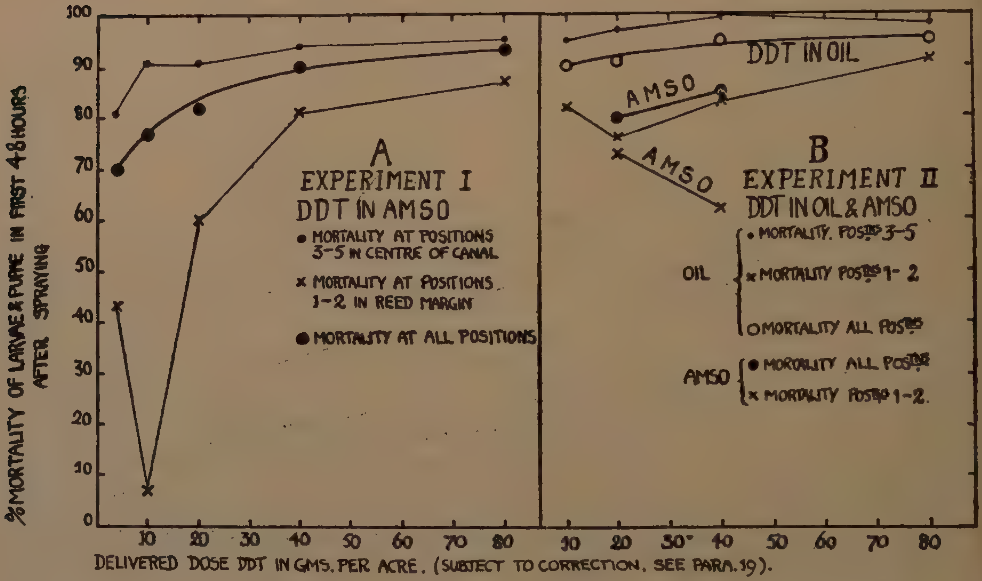

FIG. 7.—A. Experiment No. 1. AMSO; percentage mortality of larvae and pupae in the first 48 hours after spraying.

B. Experiment No. 2. DDT in oil and AMSO; percentage mortality of larvae and pupae in the first 48 hours after spraying.

Consideration of initial kill.—If figs. 7 A and B, showing the percentage mortality in the first 48 hours after spraying are compared it will be obvious that :—

(*a*) 10 and 20 g. of DDT in oil give a higher mortality than the same doses in AMSO. This superiority is seen not only in the experiments carried out with the oil and AMSO simultaneously as 2nd experiment (fig. 7 B) but also when the DDT in oil is compared with DDT in AMSO (First expt.: fig. 7 A *cfd.* with fig. 7B). The results at 40 and 80 g. DDT per acre also show a superiority of the oil over the AMSO although the differences are not so marked as at the lower dosages.

(*b*) The superiority of the oil spray is seen clearly in the kill obtained in the reedy margins (Positions 1 and 2. Figs. 7 A and B).

This higher kill with DDT in oil in the reedy margin may be due to a more satisfactory penetration of the oil compared with AMSO on the water surface in between the reed stems from spray deposited at the outer edge of the reed margin.

The generally higher kill with the oil will, at any rate in part, be due to the slightly greater amounts of DDT inadvertently delivered (as described above, p. 413). But it would seem from a comparison of the graphs that a 25 per cent. increase in dosage of DDT is unlikely to account for the much higher mortality that is shown in the reedy margins with the oil spray (see also p. 427).

Another factor to be considered is the slightly more open type of vegetation in the margins of the 2nd experimental areas. This would favour a higher mortality whatever the spray. A comparison of results for AMSO in the first and second experiments does not indicate, however, that this was of great significance (*cf.* AMSO in figs. 7 A and B).

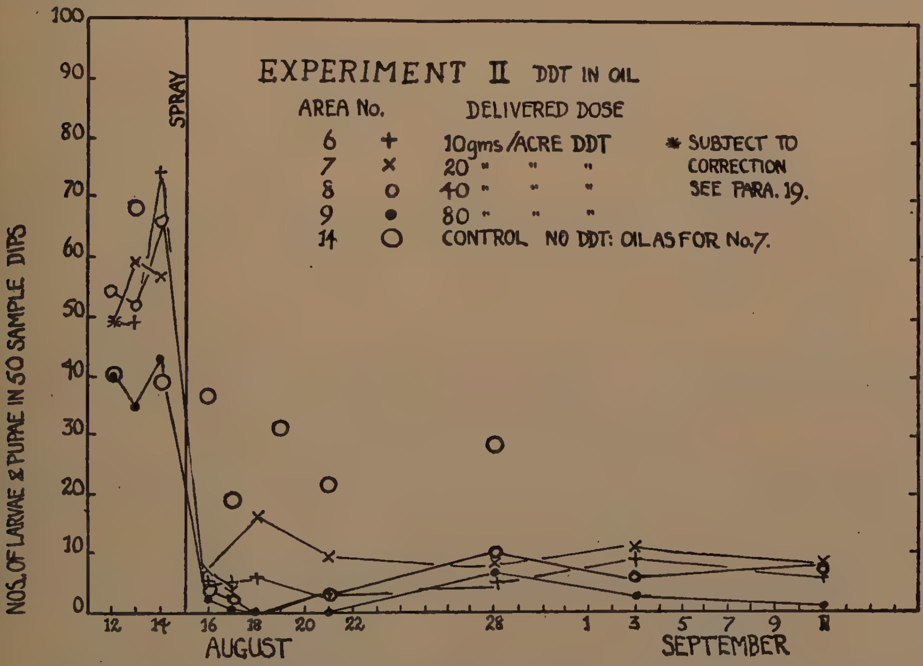

FIG. 8.—Experiment No. 2. DDT in oil. Larval and pupal counts in treated and control areas.

The population after initial kill.—If the graphs in figs. 8–10 (oil and AMSO, 2nd experiment) are compared with those in fig. 4 (AMSO 1st experiment) it will be seen that in the second trials both with oil and AMSO (figs. 8 and 10) the sharp recovery in the larval population so characteristic of the first AMSO experiments (fig. 4) does not occur to the same marked extent. A consideration of the control areas (figs. 6 and 8) bears out the suggestion put forward on p. 417 that the recovery in the first AMSO experiments was due to a general tendency for the mosquito population in the canal to increase at that time. In the 2nd experiment, the control (fig. 8) shows only a slight increase between 16th and 18th August and thereafter there is a general decline in numbers. This downward trend is shown also in the controls after 16th August in fig. 6 and in both cases corresponds to a general drop in the temperature (fig. 2). The failure of areas in the 2nd experiment to show a recovery is probably due to a general, natural decline in the population.

It is very interesting to note that with a declining population, as compared with one on the increase, the kill with a small dosage is more marked and a small dose may be almost as effective as a large one. If the population tends to increase this is not the case (*cf.* figs. 5 and 9).

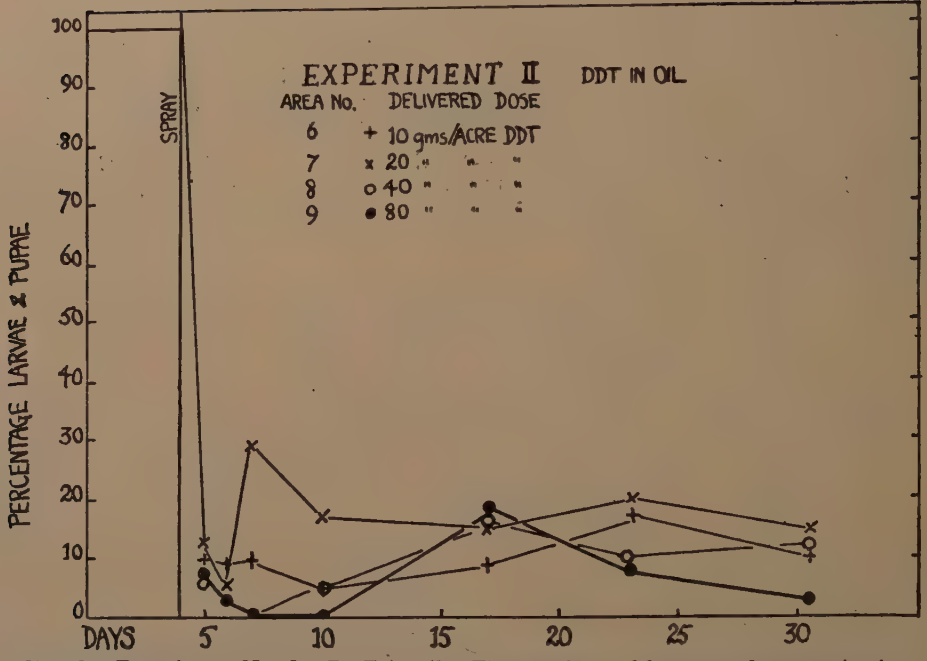

Fig. 9.—Experiment No. 2. DDT in oil. The numbers of larvae and pupae (as in Fig. 8) after spraying expressed as a percentage of the daily average of the three successive days before spraying.

To the above results can be added those obtained from two other areas sprayed at a later date (Nos. 15 and 16 of 4th experiment), where the superiority of DDT in oil as against DDT in AMSO was demonstrated.

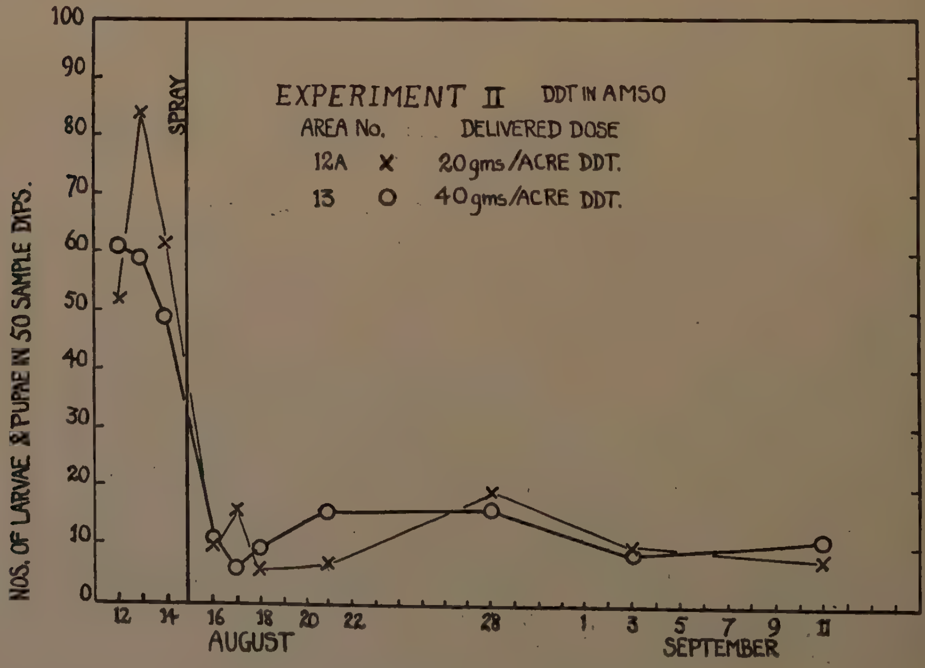

Fig. 10.—Experiment No. 2. DDT in AMSO.

Early in September it was impossible to find areas with a sufficient larval and pupal population extending right across the canal and it was therefore necessary to work entirely on the population in the reedy margin. A full description of the tests follows on pp. 423–424. It is sufficient here merely to refer to the effect on the marginal population of a delivered dose of DDT at 10 g. per acre in oil and in AMSO. From the graphs (fig. 13) it will be seen that DDT at 10 g. per acre in oil gave a substantial kill during the three days after spraying, whereas in the area sprayed with DDT at the same rate in AMSO the population actually increased. There may in fact have been a certain mortality with the AMSO since there was a much greater increase in numbers in the control areas. But the success of the treatment with AMSO bears no comparison with that of oil and in this experiment there was no excess of DDT sprayed inadvertently as had happened previously.

Third Experiment.

The effect of droplet size with DDT in oil.

Oil solution concentration	4·3 per cent. DDT.
Delivered dosages	10 and 20 g. DDT/acre.
Drop size, small	0·41 mm. diameter.
Drop size, large	0·95 mm. ,,

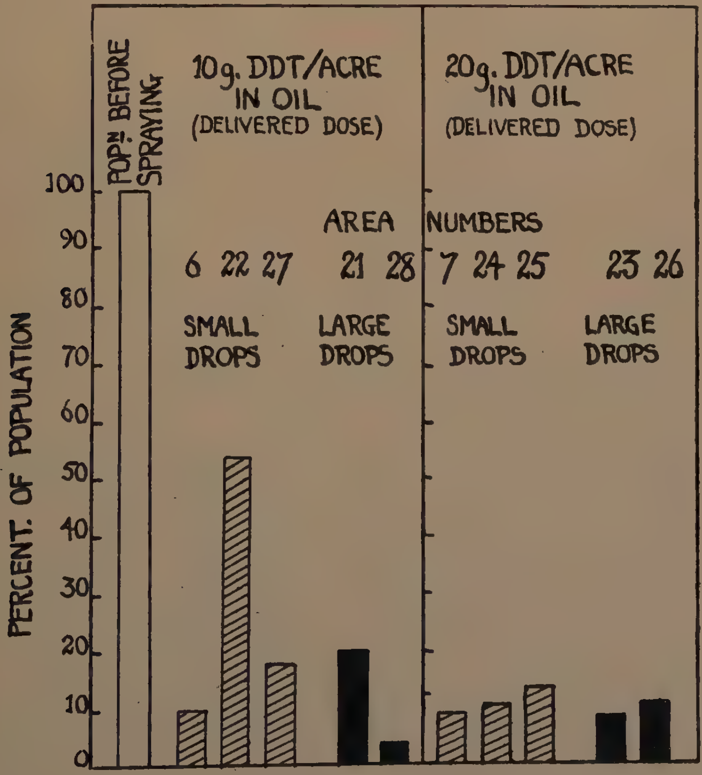

Fig. 11.—Experiment No. 3. The effects of droplet size. Percentage of population (larvae and pupae) remaining in first 48 hours after spraying. Percentage is based on mean of three days' counts before and two days after spraying.

It was thought that small drops might penetrate the vegetation or hit the small interstices of water surface between floating leaves more frequently and thus give a higher kill than large drops. For, although the diameter of the small drops was only half that of the large ones, the relative numbers were approximately 8 to 1.

Duplicate plots were selected since by this time larval and pupal counts were rather low on the remaining sections of the canal. The results are given in Table I and figs. 11 and 12.

With a delivered dosage of 20 g. per acre the results were very consistent on all four plots and drop size was without effect. This is shown clearly in the mortality for the first 48 hours after spraying (fig. 11). The similarity of all the graphs for the general level of population over the succeeding days is also striking (fig. 12).

At a delivered dosage of 10 g. of DDT per acre differences in results with two different drop sizes are scarcely more noticeable. For the higher initial mortality with the spray of larger drops is associated with higher kill in one experiment although there is little to choose between the results in the other four (fig. 11). If the initial kill for the first *three* days is taken both the areas sprayed with large drops have a higher mortality than those sprayed with small drops. The results are seen also in the population graphs (fig. 12) where a larger drop-size gives a better kill in both experiments.

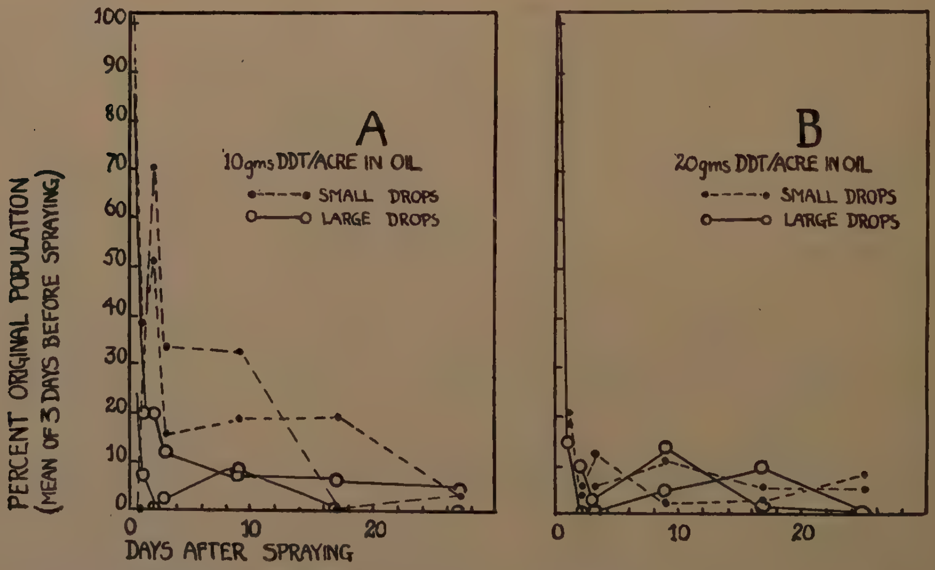

FIG. 12.—Experiment No. 3. The effects of droplet size. Population during the weeks following spraying with DDT in oil.

It is known (p. 413) that the loss of spray by slight breezes during spraying is more pronounced with small droplets than with larger ones and that consequently a smaller quantity of spray reaches the water surface if the droplets are small. This would tend to make itself shown when the dosage delivered is sufficiently low (*e.g.* 10 g. DDT per acre). With a higher rate of DDT delivery evidently enough can get down on to the water (under the conditions of these experiments) always to produce the same general level of mortality.

Fourth Experiment.

Effect of dilution of the spray liquid.

Oil solution concentrations	4·3 per cent., 0·43 per cent. and 0 per cent. DDT.
AMSO concentration	0·43 per cent. DDT.
Delivered dosage, oil and AMSO ...	10 g. DDT/acre.
,, ,, pure oil, no DDT	As for 0·43 per cent. solution, 2300 ml./acre.
Drop size	0·95 mm. diameter (large).

TABLE I.

DDT in oil. The effect of droplet size. See figs. 11 and 12.

Area No.	22	27	21	28	24	25	23	26
Dosage DDT/Acre	10 gms.		10 gms.		20 gms.		20 gms.	
Droplets	small		large		small		large	
Day	Total larvae and pupae in 50 sample dips							
1	34		31		43	49	19	65
2	26	39	52	49	28	36	28	43
3	33	24	35	59	39	23	17	37
4 sprayed								
5	12	0	8	3	7	7	3	7
6	22	11	8	0	1	2	0	5
7	5	7	5	1	5	2	0	1
13	6	7	3	3	1	4	1	7
21	6	0	3	0	1	2	2	1
29	1	1	2	0	3	2	0	0
Mean of 2 days after spraying as percentage mean of 3 days before spraying								
	55	17	20	3	11	4	7	12

Owing to a general decrease in larval numbers towards the end of the season it was necessary to work entirely in the reed margin (Positions 1 and 2). In order to increase the accuracy of the method, strips along the canal side of twice the usual length (*i.e.* 40 instead of 20 ft.) were marked out and 40 dips were taken along this length instead of 10 per 20 ft. In order to avoid undue disturbance of the water surface by dipping too closely, 20 dips, approximately 2 ft. apart, were taken along the whole length of the plot at each position. This was then repeated immediately so that each plot was sampled with 80 dips.

In order to avoid undue leakage of the dose from the margins outwards due to spreading of oil, the whole width of the canal was sprayed, although samples were taken only from the reedy margin (Positions 1 and 2).

All areas were sprayed at a delivered dose of 10 g. DDT per acre since it was thought that an effect due to dilution most likely would be seen if there was an incomplete kill.

Area 15 was sprayed with DDT in oil at the original concentration, *i.e.* that used on all other plots previously sprayed at this dose.

Area 19 was sprayed with the same amount of DDT but in ten times as much oil.

Area 17 was sprayed with the same amount of oil as in Area 19 but without DDT (Oil Control).

Area 18 was left unsprayed as a blank control.

Area 16 was sprayed with AMSO in original dilution.

Consideration of Fig. 13 shows clearly that :—

(*a*) Following spraying there was a similar increase in population in both controls and that oil alone even in ten times the quantity as in the original spray appears to be non-lethal.

(*b*) The rise in the area sprayed with AMSO is not so great as in the control areas and there has probably been a mortality. This is evidently very small, however.

(*c*) Both areas sprayed with oil solution show a very much higher mortality than the AMSO area.

(*d*) Although the difference in mortality in the two areas sprayed with DDT in oil at different dilution is not great and cannot be definitely attributed to dilution of the spray, the smaller quantity of more concentrated solution is, if anything, slightly more effective than the larger quantity of dilute spray. This may be due to a greater loss of oil and DDT because the larger quantity would tend to spread over a wider area than the smaller amount of more highly concentrated spray.

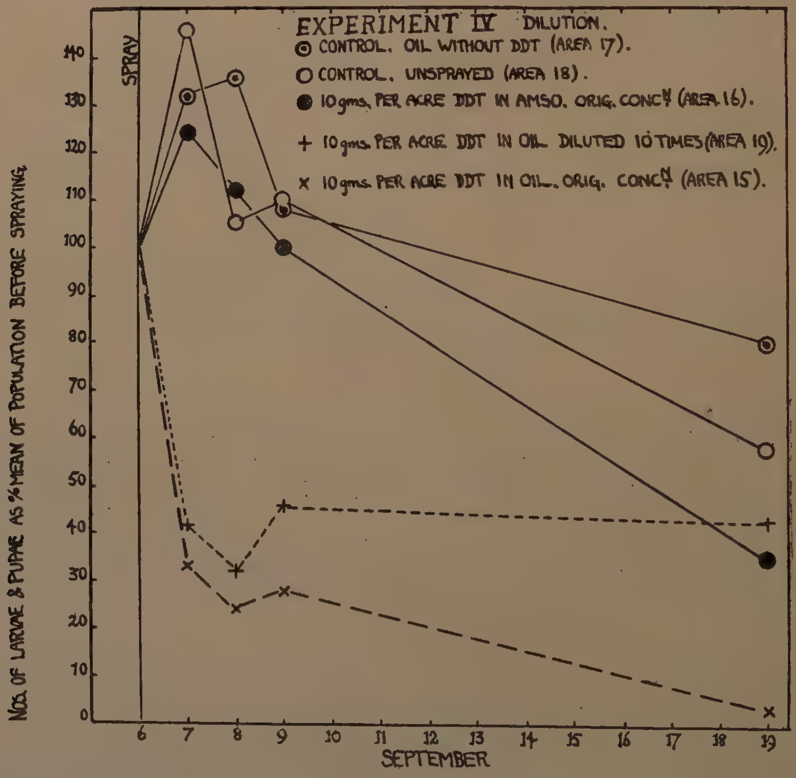

FIG. 13.—Experiment No. 4. The effects of dilution of spray. Population after spraying expressed as a percentage of original population (mean of three days before spraying).

It may be supposed that even if there was not such a loss, much of the DDT in the more dilute spray would be in the upper layers of a thick oil film and therefore fewer molecules would be available to the mosquito larvae coming up beneath.

The susceptibility of the different developmental stages to DDT and its bearing on residual effect.

There is the possibility that DDT affects the larvae by delayed action so that, although they hatch from the egg, they fail to develop beyond the third larval stage.

While it is difficult to investigate this matter with the present data and in view of the general decline of the population, it can be stated that :—

(*a*) at least some 3rd instars and later stages do develop subsequently to DDT treatment.

(*b*) That the first two larval stages are more easily killed than the 3rd and 4th stages and also more easily killed than the pupae.

(*c*) Although it is not known what proportion of later stages do succeed in developing in spite of DDT previously put down it seems unlikely that there is a strong residual effect. For although there is a general decline of the population, 3rd instars and later stages do develop and the population eventually reaches its original composition, though not its original level, about three weeks after treatment. This would be expected if there was no residual effect.

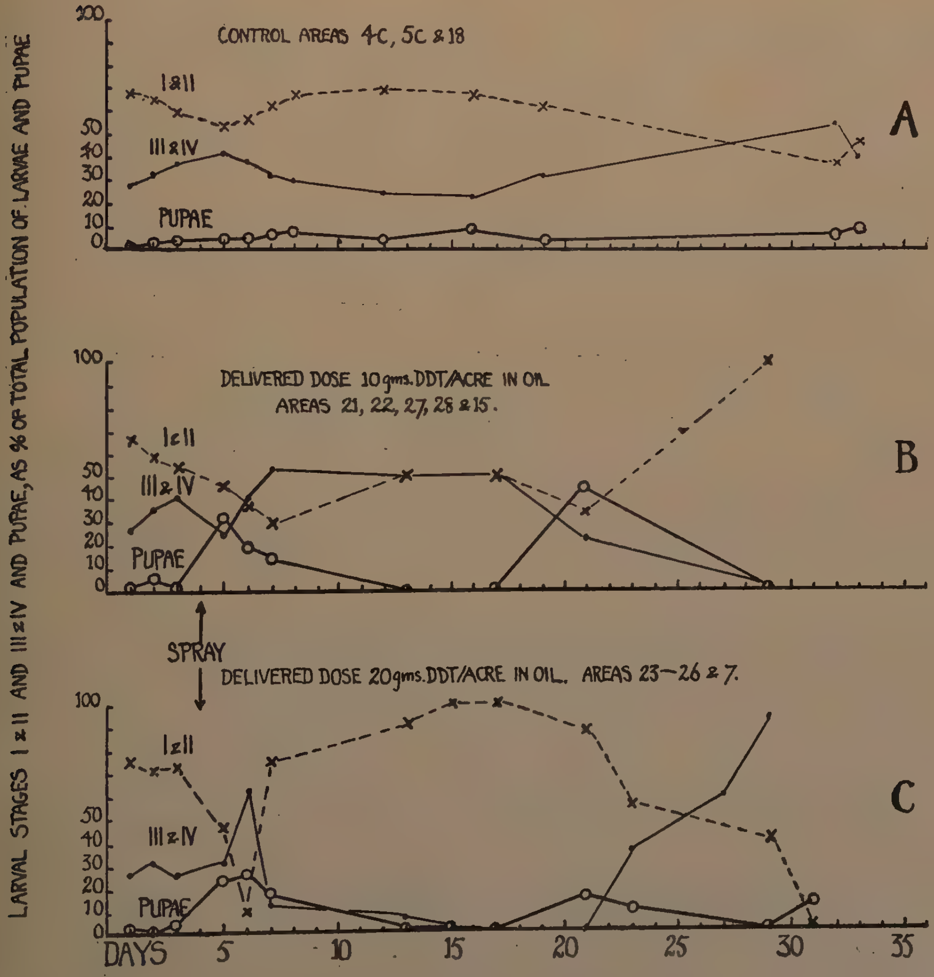

FIG. 14.—Proportion of larval instars (I & II and III & IV) and pupae in the populations before and after spraying.

Each treated and control area has been analysed in the following manner. The 1st and 2nd instars were grouped, the 3rd and 4th instars grouped and the total, together with the total pupae, were tabulated for each sampling day. The proportion of 1st and 2nd, 3rd and 4th and pupal stages in the total count was calculated and the figures graphed. This gives *not the rise or fall* of the population but the proportion of the different stages in it before and after spraying. Actually the *total* population was usually falling.

Since all areas treated with DDT gave exactly similar graphs, only a few selected ones and graphs of the totals for several similar areas are shown in figs. 14 and 15.

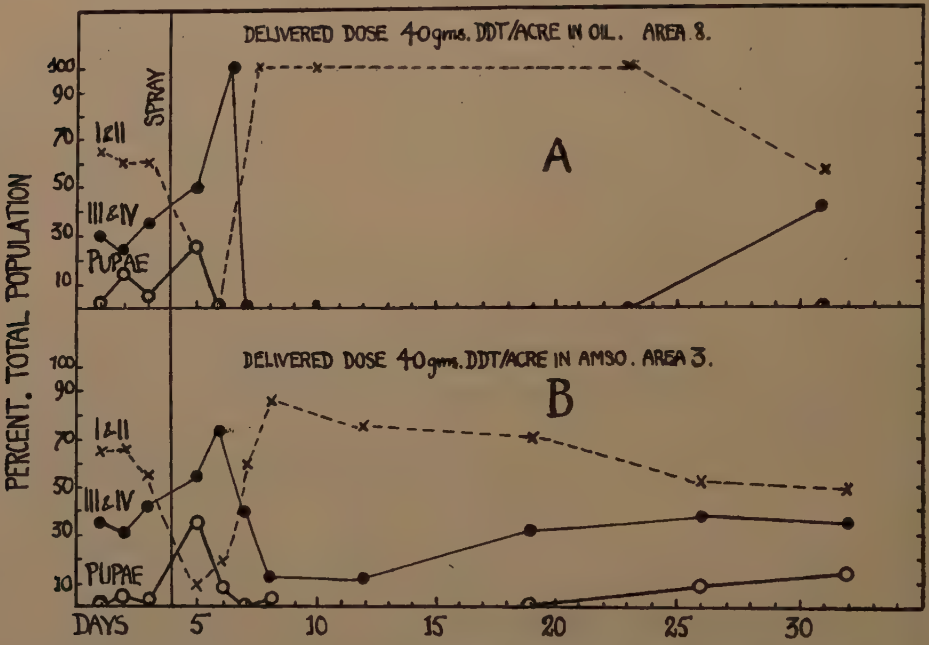

FIG. 15.—Proportions of larval instars and pupae in the populations before and after spraying.

Fig. 14A, where the control (untreated) areas are analysed, shows that the proportion of 1st and 2nd, 3rd and 4th and pupae remain fairly constant over a long period and that the early stages predominate.

When an area is sprayed (fig. 14 C) the first two stages suffer a higher mortality compared with the later larval stages and the pupae, and the proportion of pupae and later instars thus rises sharply. Soon after spraying, however, the proportion of early stages recovers and after about three weeks again declines because the young are developing into the later instars: these later stages show a corresponding increase. This is well seen in fig. 14 C and also in fig. 15. It occurs with DDT either in AMSO or in oil and it happens at all dosages.

It is suggested that if a fairly strong residual effect were present the alteration in proportions of different stages following spraying would be maintained—instead of reverting to the proportions before spraying.

To these data may be added some observations made on the persistence of oil films. It was often seen in the areas which had received a delivered dosage of 10 g. per acre of DDT in oil that the oil persisted for as long as three weeks after spraying.

This was seen particularly close to the bank (Position 1). For example, in Area 22 oil was still evident at the extreme edges of the canal among the reeds seventeen days after spraying. This oil occurred in patches of varying size up to 3–4 in. across, each patch having an irregular but discrete edge. The film therefore did not break into lenses—or if lenses did exist they were exceedingly small and inconspicuous. It was common, moreover, to find larvae of all stages in this position, so that when ladling out a sample both oil and larvae would be collected together. Over the seventeen days subsequent to spraying, 34 larvae and six pupae were collected from Position 1 in Area 22. These larvae must have been in contact with the film at some time during this period yet they were apparently tolerant of it at least after many days. This suggests that the film loses its insecticidal potency although considerable quantities of oil remain. Whether the DDT available to the larvae is reduced by spreading of the film to a point where it is rarely encountered in a sufficient concentration or whether the DDT itself deteriorates is not known.

Discussion.

The objects of the tests were to determine :—

(i) The minimum lethal dose (MLD) of DDT in oil and in AMSO.

(ii) The effects of drop size.

(iii) The effects of sprays of different dilutions in relation to amount of spray delivered.

For ease of discussion some factors can now be eliminated fairly easily. Except for very small dosages (<10 g. DDT per acre), drop size within the range 0·4 mm. to 1 mm. diameter is of no account under the conditions of these experiments and with the same doses a large number of small drops is no more effective than a small number of large ones in covering the water surface between vegetation. With solutions of DDT in oil the concentration of the DDT in solution appears to be a minor matter as far as larvicidal action is concerned provided the surface dose of DDT is the same.

We are left therefore with a discussion of the MLD of DDT in AMSO and in oil. At the same time we may discuss the assessment of actual surface dosage in relation to the delivered dose.

A consideration of the MLD with oil solution and AMSO, account being taken of the actual surface dosage in relation to the delivered dosage.

It was stated on p. 413 that with small drops the surface dosage was probably about 70 per cent. of the delivered dosage, a certain loss being caused by wind. This estimate was derived from the assessment tests in the 1st and 3rd experiments (pp. 413–414). With large drops there was evidently no loss and delivered and surface dosages are the same. This is shown by the 106 per cent. recovery of delivered dose in the assessment trial (p. 414).

With AMSO the overall mortality curve (*i.e.* at all sampling positions) drops rather sharply at delivered dosages below 40 g. DDT per acre (fig. 7 A). This drop in mortality occurs most in the reed margins (which have greater cover) and least in the unobscured central regions of the canal. It would seem that to increase the delivered dosage above 40 g. per acre would be uneconomical in the use of DDT, while to drop below 40 g. per acre would definitely result in a much lower kill—particularly where there is cover, as in the reed margin. Thus 40 g. per acre may be taken as the most economical delivered dose. This is estimated at 28 g. DDT per acre surface dose, since small drops were used.

It is evident from fig. 7A and B that the tests with oil show a more satisfactory kill than those with AMSO. The overall mortality at a delivered dose 20 g. per acre on the graph (actually 25 g. per acre due to error, see p. 413), is 91 per cent. as compared with 90 per cent. at the 40 g. per acre point on the AMSO graph (fig. 7 A) Moreover the mortality at 10 g. per acre (12·5 g. per acre actual delivered dose) was also 90 per cent.

Thus for an actual *surface dosage* estimated at 9 g. DDT per acre a 90 per cent. kill was obtained in the trials with an oil solution and a 91 per cent. kill with an estimated surface dose of 17·5 g. per acre. It is true that at a delivered dose of 40 g. per acre (50 actual) a 95 per cent. mortality was obtained (25 g. delivered in oil seems to be equivalent to 40 g. in AMSO).

AMSO is definitely inferior to oil where there is reed cover as shown in figs. 7A and B and in fig. 13 where a satisfactory minimum dose is in the neighbourhood of 56 g. per acre, surface dosage (80 g. delivered dosage). It is difficult to say with certainty that this result was not due in some way to a greater amount of reed cover in the AMSO areas. But judging qualitatively, the reed cover seemed to be about the same with all plots in this trial.

The following hypotheses may be advanced tentatively to suggest why, at low area dosages, DDT is more effective when applied in oil solution than when applied in a finely dispersed oil-in-water emulsion :—

(*a*) Some of the fine oil drops of AMSO emulsion penetrate below the water surface, thus distributing some of the DDT through the bulk of water ; solutions of DDT in oil remain on the water surface.

(*b*) The laboratory tests on emulsions, which indicated them to be superior to oils, were carried out on Culicine larvae which live largely below the surface. Since *Anopheles* larvae spend a high proportion of their active lives on the water surface they may be able to pick up a greater dosage of DDT from the localised oil film than can the Culicine larvae.

(*c*) The soap and phenolic components of the emulsifier, AMOA, containing cresols and sulphonated fats may be mild repellents for the larvae, which make them seek only the uncontaminated areas when breathing at the water surface.

From the point of view of larvicidal action, the experiments have not shown that it is an advantage to distribute DDT in high dilution, either in the form of a dilute solution in oil, or in a dilute emulsion. The efficacy of DDT is primarily dependent upon the weight dispersed per unit area rather than on the means of distribution. Drop size and solution concentration should not be increased, however, to the point where the drops are so widely separated that small breeding areas may fail to be " hit ". This limiting condition is unlikely to be approached with petroleum oil solutions of DDT (Saturation concentration c. 5 per cent.) dispersed as aircraft spray. Considerations of oil and transport economy argue the use of concentrated rather than of dilute solutions.

The actual dosage to aim at will depend on the frequency with which spraying is carried out. If, as now seems possible, the residue of spray falling on to water surfaces after anti-adult spraying is used under certain circumstances as a bonus for larvicidal effects and spraying is done at fairly frequent intervals, to aim at 100 per cent. kill of larvae may not be compatible with economy in use of insecticide; a surface dosage of 7–18 g. DDT per acre may be sufficient to produce a periodic and accumulative 90 per cent. larval mortality. This view is supported by the results graphed in figs. 4, 5 and 8 where at the end of the breeding season and with a falling larval population the slightly increased kill at a high dosage may be uneconomical in view of the similarity of the subsequent low population levels at all dosages.

It must be remembered that the minimum lethal dosages discussed here refer to the dose laid down on a very narrow strip of water and that leakage from either side was relatively large, at any rate after the first day (viz. effect on controls in 1st experiment). It is, of course, probable that all the lethal effect is produced in the first few hours after spraying and that the effect of subsequent leakage by spreading from each edge of the experimental area was negligible. But it would be supposed that on a wide area where leakage was proportionately less the MLD might even be lower than that discussed here.

In all the tests of the present series, the lethal effect of the DDT was noticeably reduced after a few days. This contrasts strongly with laboratory experiments and with known data concerning the chemical stability of DDT. It is possible that leakage of the oil films from the experimental areas may have contributed to the lowering of residual effects, but, nevertheless, the observations reported on p. 426, that both visible oil films and larvae were to be found some days after spraying, strongly suggests that the DDT has been selectively removed from the oil. An absorption of DDT by vegetation is a possible explanation of the effect. Another possible source of loss is via aqueous solution into fatty parts of various organisms in the water.

The minimum lethal dose of DDT as a larvicide has not been defined exactly : but it has been narrowed down to 40 g. DDT per acre and below. The amount of DDT to use at one spraying will depend on the frequency of spraying, whether the aim is purely larvicidal or larvicidal and adulticidal. It will also depend on the vegetation and perhaps leakage of the surface film by spreading. These points still require study. The residual lethal effect of persistent oil films appears to be low. It is clear that work is required to determine how long DDT retains its potency as a larvicide on natural water surfaces or as an adulticide to emerging or ovipositing adults coming in contact with such surfaces.

Summary.

In order to assess the potential effectiveness of DDT spray against mosquito larvae living under natural conditions and to determine the characteristics of the most effective form of spray, the variables considered were, area dosage of DDT, oil solutions compared with oil-in-water emulsions, concentration of DDT in the spray liquid, and drop size.

Spraying was carried out over small (20 ft. × 40 ft.) areas of canal harbouring larvae of *Anopheles maculipennis* var. *messeae*, and *A. claviger*. The canal was densely packed with vegetation. The central channel was overgrown with *Potomogeton natans*, so dense that in some regions there was more area of leaf than of open water surface, and in places there were large masses of floating algae (a coarse species of *Spirogyra*). At the edge of the canal near the bank there was a margin of reeds averaging about 4 ft. wide, and from 1 to 4 ft. in height.

Spraying was carried out with a specially designed spinning disc atomiser giving drops of nearly uniform size and enabling drop-size and dosage delivered to be independently varied over a wide range. The range of DDT dosages investigated was from 4 g./acre to 80 g./acre, delivered in solutions or emulsions of approximately 0·5 per cent. and 5 per cent. concentration and drop sizes of 0·4 and 1 mm. diameter. Assessment of surface dosage in relation to delivered dosage, by spraying dyed solutions, showed that about 30 per cent. of the smaller drops were lost in the wind, but that there was no appreciable loss of the coarser spray.

For 5 per cent. oil solutions of DDT drop size in the range 0·4 to 1 mm. diameter had no effect on the kill, except at low dosages (<10 g. DDT/acre) when the greater wind loss of small drops produced variable results. Tests on the effect of drop size were not carried out with emulsions.

From the point of view of larvicidal action, the experiments have not shown that it is advantageous to distribute DDT in high dilution. 4·3 per cent. DDT oil solution sprayed at 10 g. DDT/acre actually gave a slightly higher mortality than

0·43 per cent. solution at the same dosage area of DDT, but the difference may not be significant owing to uncontrollable variables. Under the conditions of these trials, both oil solutions were superior to 0·43 per cent. AMSO emulsions. Experiments on the effect of concentration at constant area dosage were made only in the reed margin of the canal and data were not obtained for open water.

In the centre of the canal, mortalities greater than 90 per cent. were obtained for both oil solutions and AMSO emulsions at delivered dosages greater than 10 g. DDT/acre, the oil solution being slightly superior. In the reed margin the kill dropped sharply with AMSO emulsions at dosages below 40 g. DDT/acre, the fall-off was much less marked with oil solution. From considerations of the average mortality in all areas, a delivered dosage of 40 g. DDT/acre (small drops) or estimated surface dosage of approximately 30 g. DDT/acre, appears the optimum for economical usage of DDT.

When areas were sprayed the first two larval stages suffered a higher mortality compared with the later larval stages and pupae, and the proportion of pupae and later instars thus rose sharply.

There was no evidence that residual lethal effects of oil films extended beyond three days. The initial kill in these trials was followed by a very low population level, due, it is thought, to a natural decline in the reproductive potential of the population.

Acknowledgements.

This work was made possible by the collaboration of many people all of whom we thank. One of us (W. H. W.) was responsible for the design of equipment and the application of the spray. The other (C. G. J.), was responsible for the entomological side of the work. The investigation was suggested by Professor P. A. Buxton, F.R.S.; the scope and broad details were planned in discussion with Drs. G. S. Hartley and W. A. Waters (Ministry of Supply). The latter also participated actively in the earlier experiments.

The work was carried out while one of us (C. G. J.) was in the Army: acknowledgements are made to the Ross Institute of Tropical Hygiene for the grant enabling the work to be prepared for publication. The work is published by permission of the Director General of Scientific Research, Ministry of Supply.

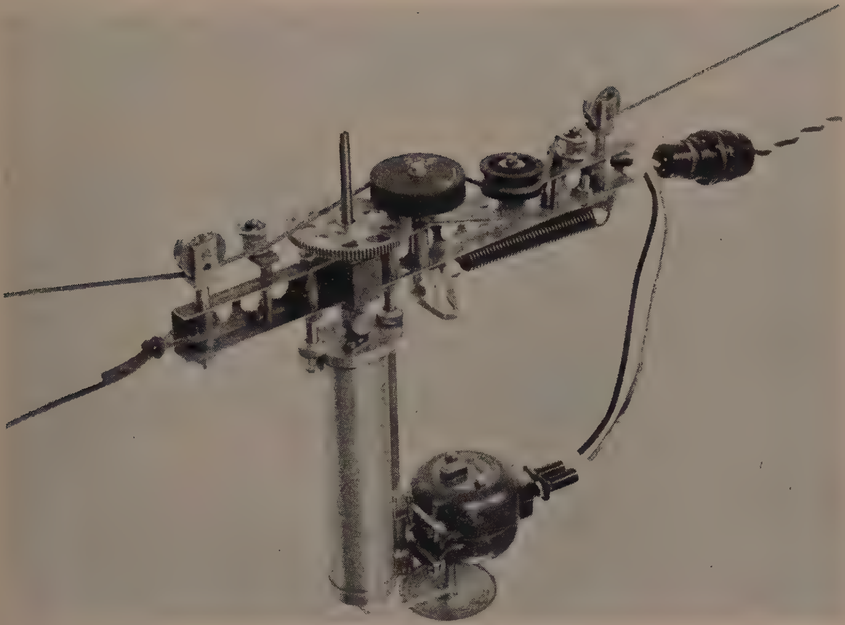

Fig. 1. The Spray Apparatus.

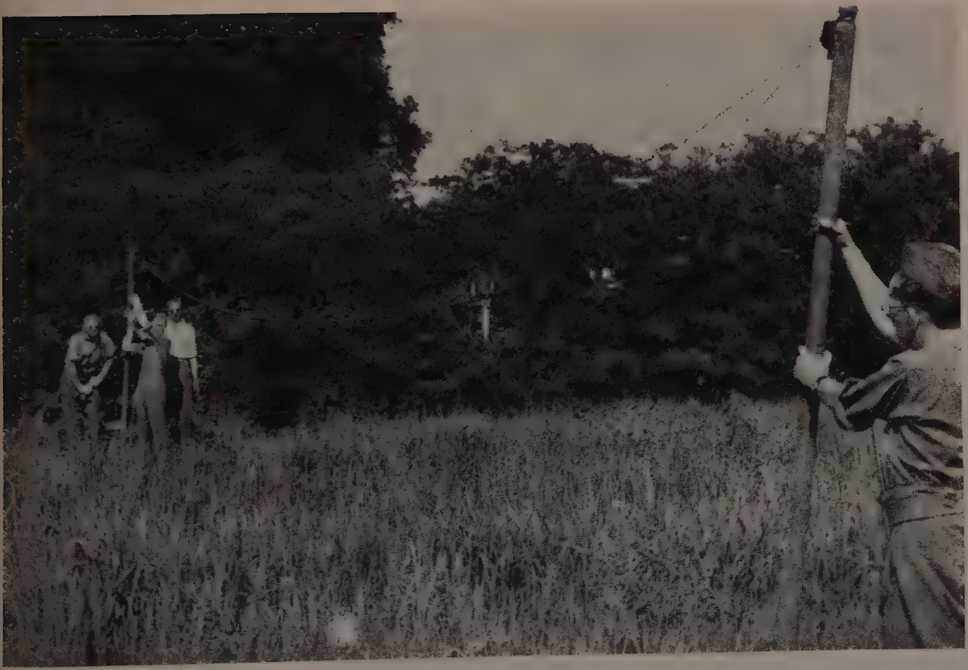

Fig. 2. The Apparatus in use.

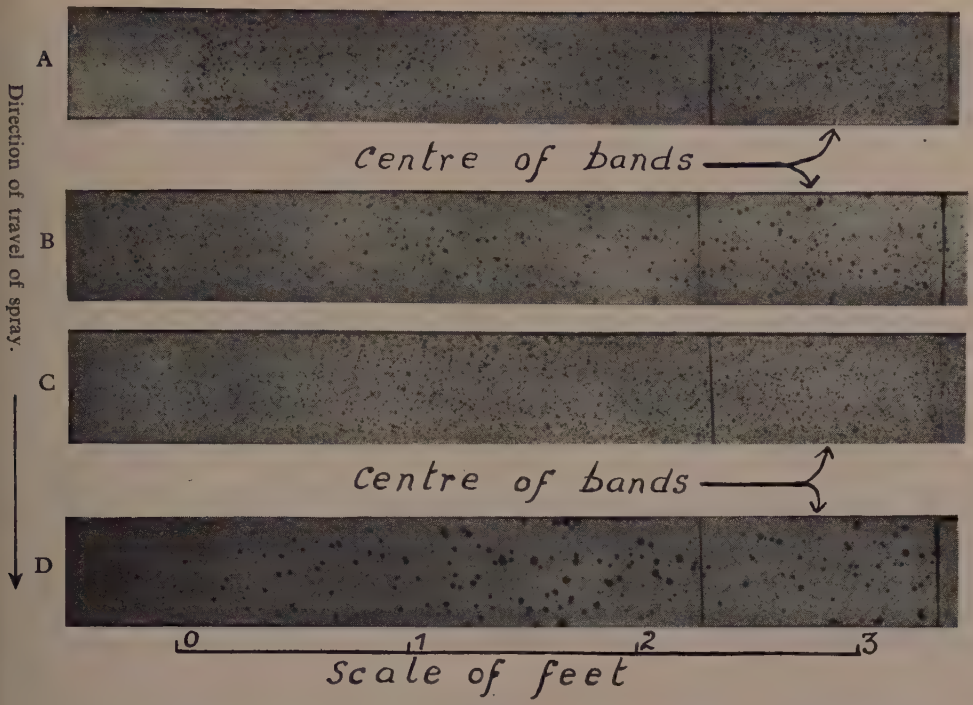
Direction of travel of spray.
A
B
C
D
Centre of bands
Centre of bands
0
1
2
3
Scale of feet

Fig. 1. Sections of sprayed bands (outside edges to the left). Dyed oil solution. Spraying at rate equivalent to 20 g. DDT/acre from 5% (approx.) solution. For explanation see Description of Apparatus in text.

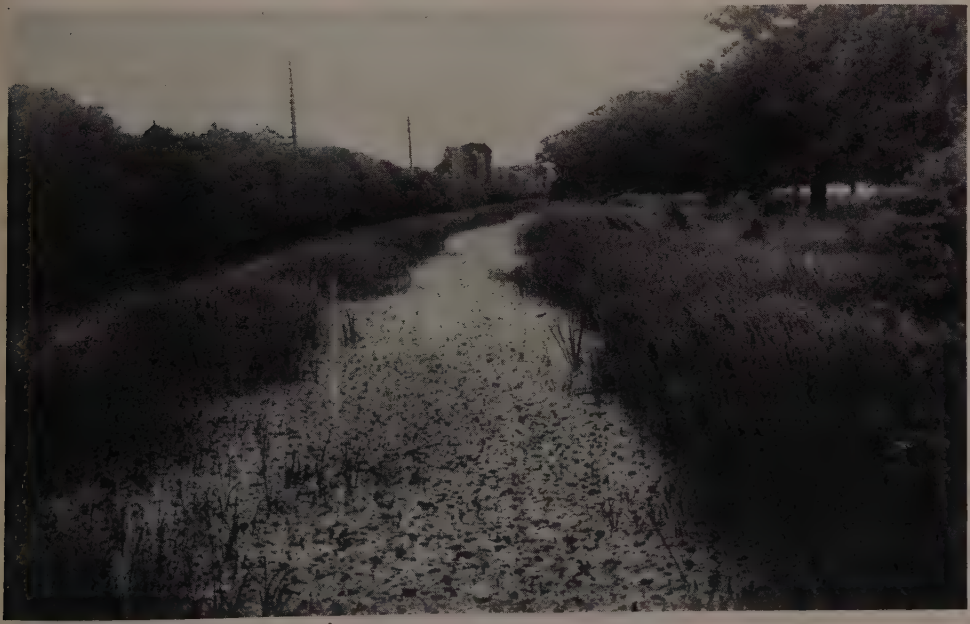

Fig. 2. General view of Canal.

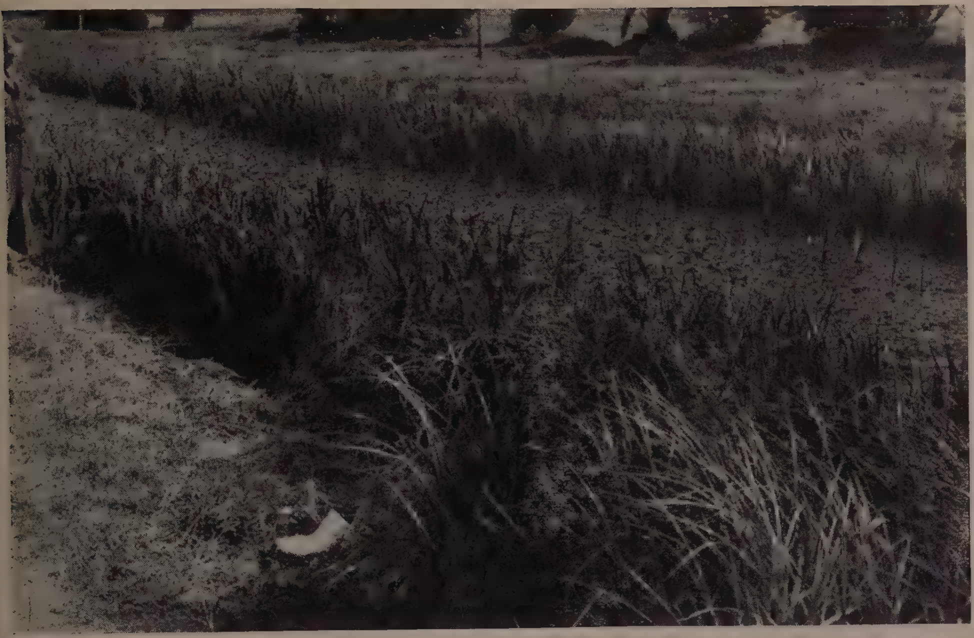

Fig. 1. Area 1c. The plot lies between the indicated points on the bank.

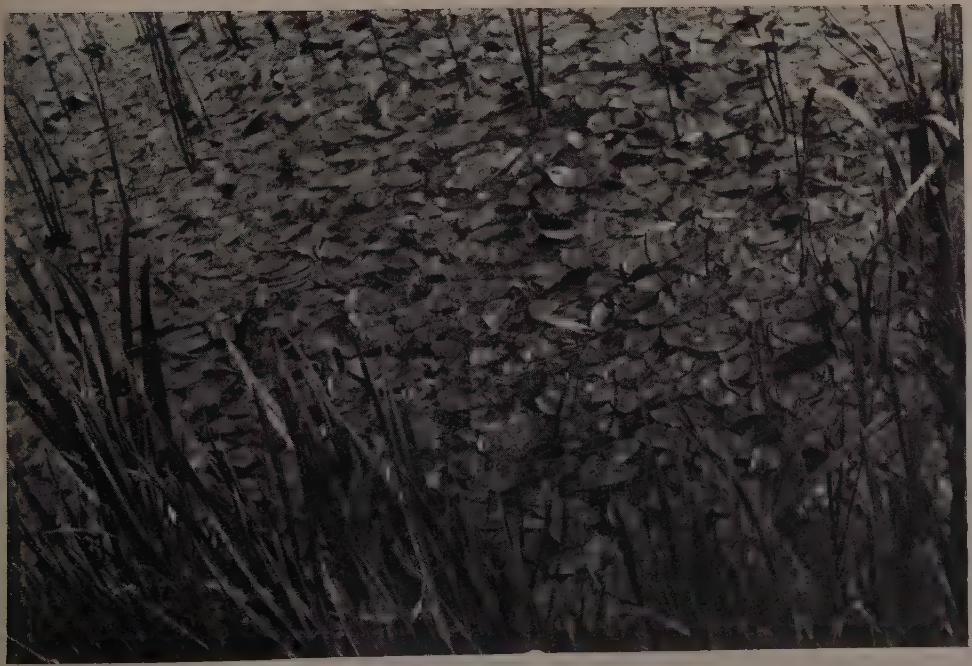

Fig. 2. Area 5. Positions 3, 4 and 5.

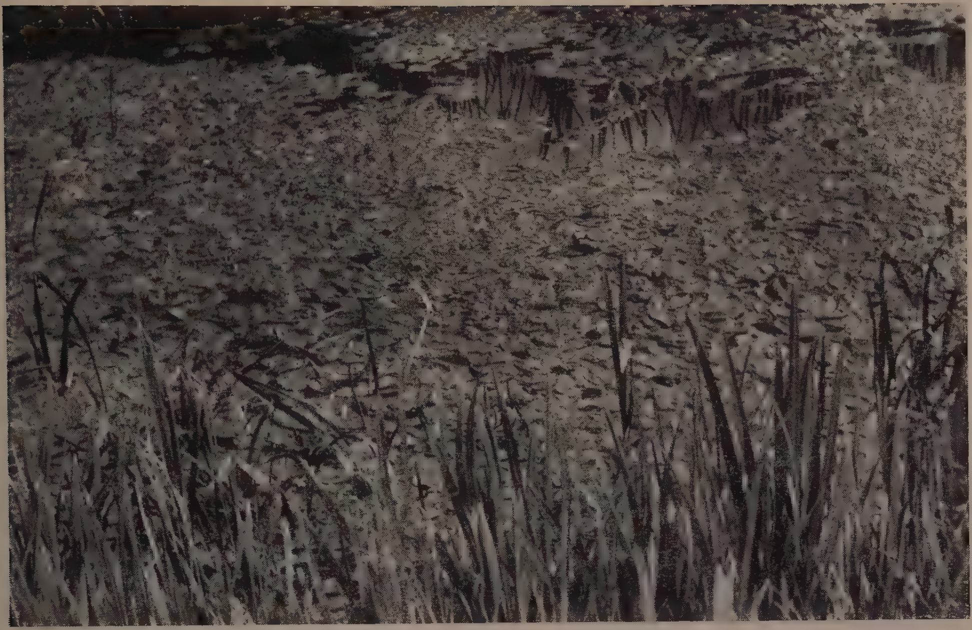

Fig. 1. Area with floating Algæ among *Potomogeton*. Open spaces as in this area were rare.

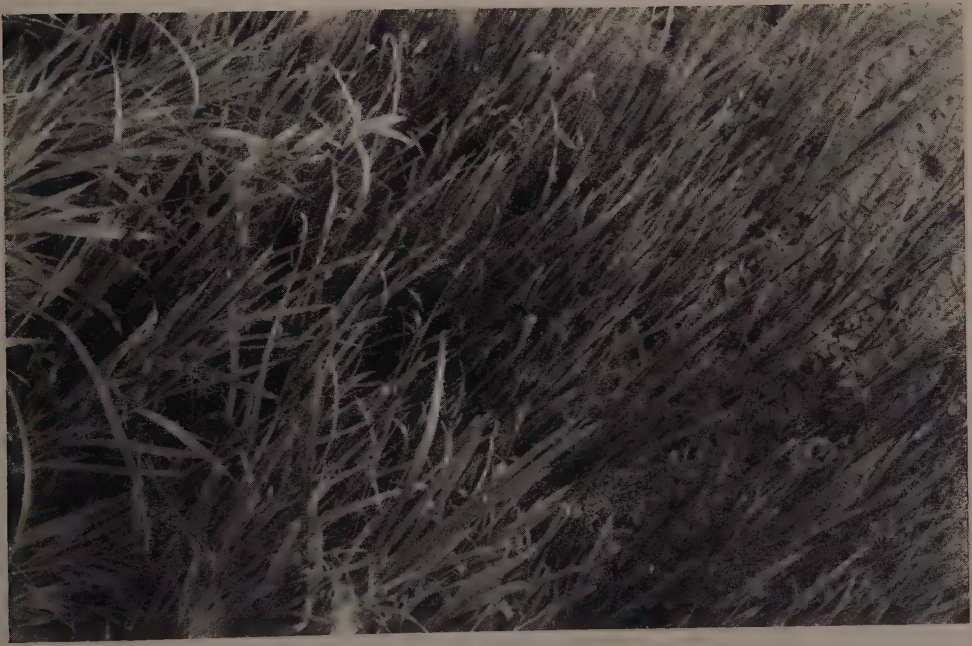

Fig. 2. Area 21. Reed margin. Typical density.

SOME OBSERVATIONS ON THE HUNGER-CYCLE OF THE TSETSE-FLIES *GLOSSINA SWYNNERTONI* AND *G. PALLIDIPES* (DIPTERA) IN THE FIELD.

By F. L. Vanderplank.

Research Officer (Entomologist) Tsetse Research Department, Tanganyika.

L.C.

During 1940 and 1941 I carried out some field experiments to test whether the mean hunger-cycle (Jackson 1933) of the tsetses, *Glossina swynnertoni*, Austen, and *G. pallidipes*, Austen, was related in any way to the climatic conditions. Owing to the pressure of other work these experiments had to be discontinued, but it was hoped to complete them at a later date. In April 1943 a small " check " experiment on the same lines was carried out but, as I have now returned to England, it is unlikely that it will be possible to complete this work. The data obtained is of value to other tsetse workers and entomologists, particularly in connection with the recent development of new insecticides, so it is thought desirable to publish the results without further delay.

The experiments were carried out in a " hardpan " area in experimental Block 10C at Old Shinyanga, Tanganyika. This area has already been described in a previous paper (Vanderplank 1944).

Experimental Technique.

Only male *swynnertoni* or *pallidipes* were used in these experiments and were caught off a tethered ox. They were hunger-staged according to external appearance (Jackson 1933) and their proboscidae were cut off with a pair of fine scissors, taking care not to damage the fly or its antennae in any other way. Two hunger-stages, 1 (replete) and 4 (very hungry) were treated in this manner, being marked with the date and distinctive colours for their hunger-stage and then released. A percentage of those marked were recaptured on subsequent days; these recaptures were hunger-staged, recorded, then killed and brought into the laboratory where they were checked under the microscope to make quite sure that their proboscidae had been properly cut. Only one was rejected on these grounds.

The number of marked tsetses recaptured varied between 15 and 30 per cent. This percentage is not affected by the season or the hunger state of the fly when marked but is dependent upon the density of the population, number of baits used and time employed for recapturing; also, to a smaller extent on the colours with which the tsetses are marked. Since no attempt was made at estimating the populations at different seasons, the numbers recaptured each month are not comparable, but this does not alter the validity of the estimated hunger-cycle. It should also be noted by readers who are not acquainted with tsetse-flies in the field that a bait attracts many male flies that do not necessarily want to feed, but are actively searching for females. Female tsetses on the other hand are active during their first few days of adult life but after the 7th or 8th day, when pregnancy commences, they only appear when requiring food. On account of this, population and other estimates which make use of random sampling are based entirely upon catches of the male tsetses.

Results.

G. swynnertoni.

Table I shows the results obtained by this method and the numbers of *G. swynnertoni* recaptured in each case. During December 1940, January, February, June and

TABLE I.

Numbers of marked male *G. swynnertoni* recaptured.

Hunger-stage released and recaptured	Interval between release and recapture in days												
	1	2	3	4	5	6	7	8	9	10	11	12	Mean
December 1940													
4 and recapt. as 4	3	12	—	—	—	—	—	—	—	—	—	—	1·80
1 ,, ,, 2	1	19	1	2	—	—	—	—	—	—	—	—	2·18
1 ,, ,, 3	—	1	4	11	6	1	1	—	—	—	—	—	4·04
1 ,, ,, 4	—	—	—	2	2	4	5	5	—	3	2	—	7·35
January 1941													
4 and recapt. as 4	—	8	—	—	—	—	—	—	—	—	—	—	2·00
1 ,, ,, 2	—	11	—	2	—	—	—	—	—	—	—	—	2·30
1 ,, ,, 3	—	—	1	6	6	1	1	—	—	—	—	—	4·67
1 ,, ,, 4	—	—	—	2	—	3	3	6	—	3	1	1	7·78
February 1941													
4 and recapt. as 4	—	16	14	4	—	—	—	—	—	—	—	—	2·94
1 ,, ,, 2	—	2	6	1	—	—	—	—	—	—	—	—	2·89
1 ,, ,, 3	—	7	6	2	5	2	2	—	—	—	—	—	3·79
1 ,, ,, 4	—	—	—	—	4	3	7	2	3	1	—	—	7·00
March 1941													
1 and recapt. as 2	—	—	3	—	—	—	—	—	—	—	—	—	3·00
1 ,, ,, 3	—	—	2	—	2	4	2	—	1	—	—	—	6·64
1 ,, ,, 4	—	—	—	—	2	4	5	10	10	5	4	—	8·32
April 1941													
1 and recapt. as 2	1	1	—	—	—	—	—	—	—	—	—	—	1·50
1 ,, ,, 3	—	—	—	2	—	1	1	—	—	—	—	—	4·25
1 ,, ,, 4	—	—	—	—	—	1	4	4	1	—	—	—	7·50
May 1941													
1 and recapt. as 2	6	3	2	—	—	—	—	—	—	—	—	—	1·64
1 ,, ,, 3	—	2	4	4	7	3	2	—	—	—	—	—	4·50
1 ,, ,, 4	—	—	—	—	1	3	16	9	1	—	—	—	7·23
June 1941													
4 and recapt. as 4	22	—	—	—	—	—	—	—	—	—	—	—	1·00
1 ,, ,, 2	5	1	—	—	—	—	—	—	—	—	—	—	1·16
1 ,, ,, 3	—	8	4	—	—	—	—	—	—	—	—	—	2·50
1 ,, ,, 4	—	—	—	3	8	2	1	—	2	1	—	—	5·83
July 1941													
4 and recapt. as 4	29	—	—	—	—	—	—	—	—	—	—	—	1·00
1 ,, ,, 2	6	1	—	—	—	—	—	—	—	—	—	—	1·14
1 ,, ,, 3	—	9	10	1	—	—	—	—	—	—	—	—	2·60
1 ,, ,, 4	—	4	11	34	20	2	1	—	—	—	—	—	4·11

July 1941 a number of stage 4 (very hungry) flies were marked, proboscidae cut and released with subsequent recapture. The purpose of this experiment was to ascertain how long hungry flies could exist without feeding. As will be seen in the table (December 1940), three were recaptured after one day at liberty, but it is possible, of course, that these would have been able to live longer, and twelve were recaptured after 2 days, none after this. So, under the conditions prevailing in that area in December 1940, hungry flies from a random sample of the population had, at the most, two days reserves during which time they had to feed or else die of starvation. On an average they were able to find food again within 1·80 days; actually they may have found it earlier than this but being unable to feed, were unable to satisfy their hunger and continued their search for food. In January 1941 no fly was caught on the first day after release; but eight were recaptured on the second day and none subsequently. In February the majority, 16, were recaptured on the second day, but 14 also on the third and 4 on the fourth day, so it would appear that the hungry flies in February have either better reserves of food, or the conditions are more suited to their survival at that time of year.

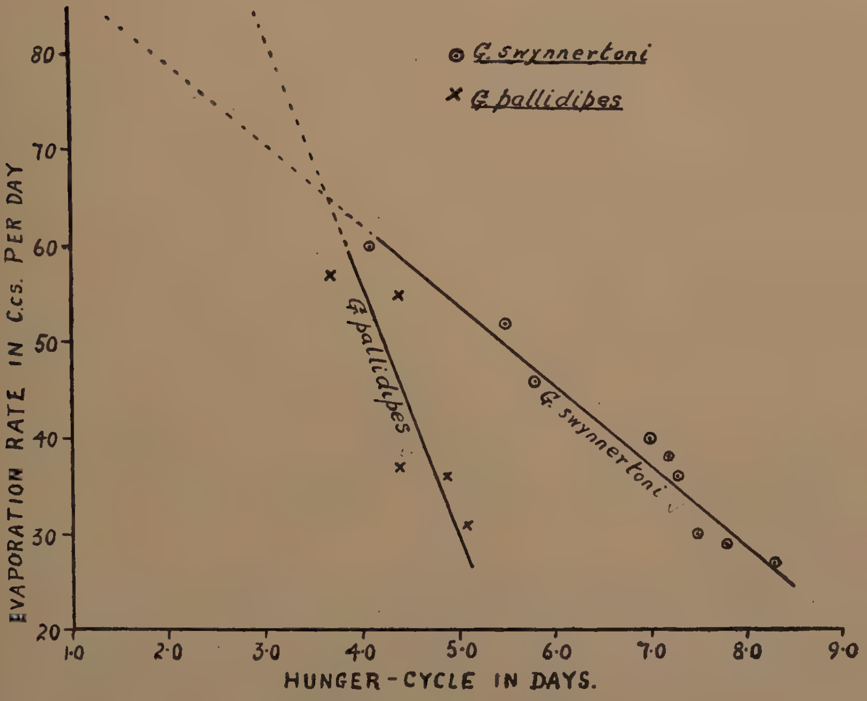

FIG. 1.—Hunger-cycle against evaporation rate of *Glossina swynnertoni* from Table II (1 & 2) and *G. pallidipes* from Table IV (1 & 6).

TABLE II.

Male *G. swynnertoni*.

	1940		1941						
	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	July
1. Mean hunger-cycle in days	5·5	7·3	7·8	7·0	8·3	7·5	7·2	5·8	4·1
2. Daily evaporation rate in ccs. Atmometer in Hardpan	52	36	29	40	27	30	38	46	60
3. Daily evaporation rate in ccs. at Standard Station	59	43	39	52	39	48	42	58	72
4. 2 p.m. saturation deficits monthly means, Standard Station	25·6	21·0	14·2	20·5	17·8	18·7	20·7	21·9	24·0
5. Mean maximum Temperature °C. at Standard Station	30·9	29·7	27·4	30·8	30·0	30·3	29·3	29·9	29·9
6. Mean Minimum Temperature °C. at Standard Station	19·0	17·9	17·7	18·2	18·4	18·8	18·4	17·5	15·6
7. Mean Median (½ Max.+ ½ Min.) temperature at Standard Station ...	25·5	23·8	22·6	24·5	24·2	24·6	23·9	23·7	22·8
8. Mean wind velocity at Standard Station in m.p.h.	3·5	1·1	1·6	2·4	2·1	2·7	2·4	2·8	3·8

Table II gives the observed mean evaporation rates at the catching site, and from the Standard Meteorological Station at Old Shinyanga, some three miles away, the latter being situated in cleared country ; the table also includes the mean 2 p.m. saturation deficits and the mean maximum, minimum and medium temperatures, the mean wind velocity, all recorded at the Standard Station. February, as it will be seen, was both hotter and drier than the other months, and since the results obtained for February are not in line with the other observations, it is suggested that the tsetses were generally in a better condition, with more fat reserves during this month (when a short dry break occurs during the rains) than during the preceding months. During March, April and May no stage 4 flies were marked, but in June and July all recaptures were made after one day's interval and none subsequently, so the maximum amount of fat reserves at that time did not exceed a day's supply. The evaporation rate was slightly higher in June than in February, but the temperature was lower during the former month and so no more water loss should have been experienced in June than in February and yet no tsetses were able to survive more than one day. In July climatic conditions were more severe, but many tsetses were recaptured on the first day after marking. It is not possible to compare the numbers recaptured each month with one another since these depend on the numbers marked, population level, and the number of parties engaged in recapturing. Owing to the numbers of staff that would have been required, accurate population estimates could not be made in these experiments.

The main consideration, however, is the interval of time between one meal and the next in the field. Those tsetses which were allowed to gorge before having their proboscidae cut and then marked and released, provide the main data for the determination of the mean hunger-cycle under various seasonal climatic conditions. Such flies were recaptured in different hunger stages and these are given in Table I. The time taken between a meal and being diagnosed as hungry (stage 4), that is, ready to feed again, varied between 4 and 12 days in January 1941, and between 2 and 7 days in July 1941, and can be described as the maximum and minimum observed hunger-cycles. The differences between these observed maxima and minima appear to be unduly large, but may be due to numerous causes, such as the age of the fly (young flies in the laboratory take smaller meals and digest them quickly), its activity at that time, and the size of the original meal taken. In the laboratory, temperature appears to have the biggest effect upon the duration of the mean hunger-cycle and humidity very little effect. Eventually as the tsetse gets older so the hunger-cycle becomes shorter until it averages 3 to 4 days, being practically unaffected by either temperature or humidity. The reasons for this are not known, but it is suggested that it is most probably due to the cuticle, which has been shown by Wigglesworth (1945) and Beament (1945) to be the insect's waterproofing layer, getting rubbed away in confinement. Such damage may occur naturally in the field, and it is possible that handling the tsetses may damage them to a certain extent in this manner ; so the results obtained will not represent the true mean hunger-cycle of the whole population. However, it does give an indication of the mean hunger-cycle at different seasons, temperature and humidity levels, which will be approximately representative of the true state of affairs.

The effect of the oil paints used to mark the flies has been gone into very carefully both in the laboratory and in the field. It was found that certain paints, mostly greens containing arsenic or chromium salts, were poisonous to the flies, and these were discarded. Certain yellow paints appear significantly to enhance the longevity of the fly in captivity for reasons not known. In the field some colours render the tsetses more conspicuous to predators than others—this factor varies very much with the season and the species of predator active at the time ; in some seasons and areas Jackson (1941) has found that there are no differences between tsetses marked with conspicuous colours and those with small inconspicuous marks. However, it is possible to arrange the experiments using two or three colours on each fly, so that no bias due to marking is experienced. A full account of the laboratory and field experiments with different colour paints will be given in another paper.

Table II compares the observed monthly mean hunger-cycle, based on releasing fully gorged tsetses (hunger stage 1) and recapturing a portion of them as hungry flies (hunger stage 4), with atmospheric humidity measured by evaporation rate—using a white bulb Livingstone atmometer three feet above ground level, either at the site or at the Standard Station three miles away, or with the 2 p.m. saturation deficits, temperatures, and wind velocity at the Standard Station. The mean hunger-cycle correlates significantly with each of these methods of measuring humidity, and has also been correlated significantly with other measurements of humidity such as vapour pressure, relative humidity and mean saturation deficits, the means of which were calculated on hourly observations from automatic thermohydrographs in a Stevenson Screen at the Standard Station. These were not included in the above table since they are merely redundant. These significant correlations with all measurements of humidity are to be expected since all the different methods of measuring humidity correlate with each other. It is, however, difficult to decide which method should be used for the comparison, for if either relative humidity, vapour pressure or saturation deficit are used, these fail to allow for any effects wind velocity might have in increasing evaporation at constant humidities and temperatures. All humidity measurements are affected by the air temperature, but since the variation of the monthly mean air temperature over the whole period was only 2·9°C., temperature can have had only a minor influence in these observations. The obvious comparison is then between the observed hunger-cycle and the evaporation rate recorded at the experimental site. These figures give a significant correlation $r=-0{\cdot}88$ (when $N=250$ the original individual data were correlated and not the means as given in Tables), so, as the dryness of the air increases, the hunger-cycle shortens; this is shown in fig. 1.

The evaporation rate and the 2 p.m. saturation deficits from the Standard Station are included in Table II, since if one required an indication of the length of the mean hunger-cycle it would not always be convenient to set up numerous meteorological stations in the bush, and as expected they correlate significantly with the observed mean hunger-cycle ($r=-0{\cdot}94$ and $-0{\cdot}80$ respectively). Wind velocity recorded at the Standard Station and the maximum, minimum and median ($\frac{1}{2}$ max. $+\frac{1}{2}$ min.) temperatures do not correlate with the observed hunger-cycle. It would not be expected that wind velocity had a direct effect and its effects are accounted for by the evaporation rate; temperature is known to affect the hunger-cycle but as stated above the differences between the mean monthly temperature is only 2·9°C., and is insufficient to have any recordable effect. Other work suggests that the higher the temperature the shorter the hunger-cycle.

During April 1943 an attempt was made to continue the above experiments, but owing to the pressure of other work it was only possible to mark small numbers of fully gorged (stage 1) *swynnertoni* cutting their proboscidae and releasing them during that month only. The results obtained from 5 recaptures that completed the full cycle are given in Table III, and do not depart significantly from the former observations.

TABLE III.

Numbers of marked male *G. swynnertoni* recaptured.

Hunger-stage released and recaptured	Interval between release and recapture in days										
	1	2	3	4	5	6	7	8	9	10	Mean
April 1943											
1 and recapt. as 2	2	—	—	—	—	—	—	—	—	—	1·00
1 " " 3	—	—	1	—	—	—	—	—	—	—	—
1 " " 4	—	—	—	1	—	1	2	—	1	—	6·60

G. pallidipes.

A further series of similar experiments was carried out with *G. pallidipes* in another area of the field during June to October 1940, and the results are summarised in Table IV, which gives the numbers of recaptures on which the mean hunger-cycle has been based, also the mean evaporation rates observed in the field, using an atmometer sited in a thicket bordering a dry river bed, a favoured haunt of *pallidipes*; and at the Standard Station three miles away at Old Shinyanga. The table also includes the mean weight of a random sample of hungry (stage 4) *G. pallidipes* taken at the same time. For a description of the area where these experiments were carried out, known as Experimental Blocks 4A and 4B, see Vanderpank (1944b). The observed field evaporation rate is much lower than that observed at the Standard Station due to the presence of moisture in the sandy river bed which also had an appreciable cooling effect in its immediate neighbourhood. These results have also been graphed in fig. 1, and lie along a different line from that obtained by graphing the hunger-cycle of *swynnertoni* against humidity. In calculating the degree of correlation between the hunger-cycle of *swynnertoni* and *pallidipes* and the evaporation rate, it is assumed that the points obtained lie roughly in a straight line; this assumption is most probably incorrect since the correct line would be a complex curve and influenced by factors other than humidity alone. In either case, whether we assume that the lines are straight or curved, both lines meet and intersect when the hunger-cycle of both species is about 3·4 days, and the evaporation rate between 61 and 67 ccs., per day. Jackson (1945) found that "In the *morsitans* group, smaller species and smaller individuals among teneral (unfed) flies lose practically the same amount of weight as do larger species and individuals and therefore lose more proportionally. It follows that, other things being equal, smaller individuals and smaller species are at a disadvantage. It is conjectured that *swynnertoni* counteracts this disadvantage by adapting itself to taking more frequent meals, which it is able to do because it is accustomed to live among a heavy population of Ungulate animals." The present field data, however, appear contrary to Jackson's laboratory experiments, but the latter were carried out at a relative humidity of 50 per cent. and temperature 25·3°C., which is approximately equivalent to an evaporation rate of 65, when the wind velocity is 1·5 m.p.h. If we look at the evaporation rate of 65 ccs. per day on the figure, we find that it is probable, depending whether the line is straight or curved, that *pallidipes* will have approximately the

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TABLE IV.

Male *G. pallidipes*.

	1940				
	June	July	Aug.	Sept.	Oct.
1. Mean hunger-cycle in days	5·1	4·9	4·4	4·4	3·7
2. Number of recaptures on which the mean hunger-cycle is based	9	72	56	74	28 (N=239)
3. Mean weight of hungry stage 4, including the marked recaptures	36·78	36·53	35·40	34·15	31·46
4. Number of individuals on which the means in (3) are based	100	300	100	100	100
5. Mean daily evaporation rate in ccs. of Standard Station	73	72	79	88	85*
6. Mean daily evaporation rate in ccs. of Atmometer sited in thicket in the experimental area	31	36	37	55	57* ✓

*First 16 days of October concurrent with observations before the rains broke.

same hunger-cycle as *swynnertoni* at this figure. Actually Jackson was investigating the loss of weight and not the hunger-cycle, but the two are closely associated, and although *pallidipes* is considerably larger than *swynnertoni* and able to take relatively heavier meals, it has a much shorter hunger-cycle and consequently must lose weight more rapidly than *swynnertoni*. If Jackson's experiments were repeated under different temperatures and humidities a reverse effect, in line with the above experiments, might be found.

Discussion.

In Table IV when the mean (actually using individual data so as to obviate any bias) hunger-cycle is correlated with the observed field evaporation rate $r=-0{\cdot}65$ when $N=239$ and so is very significant; similarly with the observed evaporation rate at the Standard Station $r=-0{\cdot}32$ and with the mean weights of a random sample*, actually against their own weight (each recaptured fly was immediately weighed when caught) $r=+0{\cdot}85$ both of which are highly significant. From these results it can be concluded that during the early dry season (June) the tsetses have more reserves as shown by the higher average weight of the sample and do not require to feed so frequently as they do later in the dry season (October) when they have smaller reserves and also smaller individuals appear. It is well known to tsetse workers in the field that during the hotter and drier seasons females give birth to smaller larvae, and smaller adults emerge. In this respect the field work agrees with Jackson's conclusion that smaller individuals are at a disadvantage and this disadvantage is counteracted by the tsetses taking more frequent meals.

The mean hunger-cycle must not be taken as the frequency with which a sample of the male population will be seen in the field; in itself it cannot be used as a factor for correcting the mean catch (apparent density) in order to obtain the relative population (actual density), unless one only takes into account the hungry tsetses. The male spends a short interval after the meal resting and digesting that meal, then becomes active searching for females, which are most readily found around any host. Consequently a bait-animal attracts many tsetses that are not wanting to feed. In order to estimate the frequency at different times of the year, all individuals in all hunger stages would have to be marked, have their proboscidae cut and be released. In the above experiments stages 2 and 3 have not been treated in this manner, so it is not possible to work out the mean frequency with which a section of the population becomes active; this, however, probably varies according to the hunger-cycle.

Summary.

The mean hunger-cycles of *G. swynnertoni* and *G. pallidipes* have been studied in the field by marking tsetses with cut proboscidae, which have been released and subsequently recaptured.

The length of time that hungry flies are able to survive before dying from starvation varies with the season, probably according to the amounts of reserve fats and atmospheric humidity.

The mean hunger-cycle also varies with the season and correlates significantly with the evaporation rate or other measurements of humidity. The variation of temperature experienced in these experiments was not sufficient to have any appreciable effect on the hunger-cycle.

G. pallidipes, a thicket haunting fly and larger than *G. swynnertoni*, a thorn bush ore species, feeds ~~less~~ frequently than the latter under moist conditions, but ~~more~~ /less frequently under adverse conditions.

Smaller individuals probably feed more frequently than larger individuals.

---

*NOTE.—The mean weights given in the Table have been based on a larger random sample than the recaptured marked flies, so as to be more representative of the population.

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SOME NEW AND IMPERFECTLY KNOWN GALL MIDGES (ITONIDIDAE: DIPTERA) FROM INDIA.

By M. S. Mani.

Assistant Professor of Zoology, St. John's College, Agra.

E.M.N.

The types of the new forms described in this paper are deposited in the collections of the Zoological Survey of India, Kaiser Castle, Benares.

Tribe Lasiopterini.

Prolasioptera javanica (Kieff. & Leeuw.-Reijn.).

Lasioptera javanica, Kieffer & Leeuwen-Reijnvaan, 1909. Marcellia, **8**, p. 123, fig. 10.

Prolasioptera javanica, Kieffer, 1913. Gen. Ins., **152**, p. 33.

I refer to this species the midges causing galls on stem, petiole and mid-rib of *Melothria perpusilla* Cogn. (Gall No. 225), *M. heterophylla* Cogn. (Gall No. 223) and *M. amplexicaulis* Cogn. (Gall No. 222) in South India.

Although the galls of this species were described nearly ten years ago, this is the first definite record of the species from India (Mani, 1935). The species was originally described as causing galls on *M. perpusilla* Cogn. in Java.

Tribe Oligotrophini.

Oligotrophus saligneus, Coquillett. No.

Oligotrophus saligneus, Coquillet, 1903. Indian Mus. Notes, **6** (1) p. 1.

Coquillett's original description of this species was apparently unknown to Kieffer, who in 1913 quite erroneously credited the species to Lionel de Nicéville, adding in parentheses "*sine descrip.*". This mistake was repeated by Senior-White (1928) and other workers. I have recently discovered Coquillett's description, which is valid under the International Rules of Zoological Nomenclature. Coquillett is therefore the author and not de Nicéville as cited in literature.

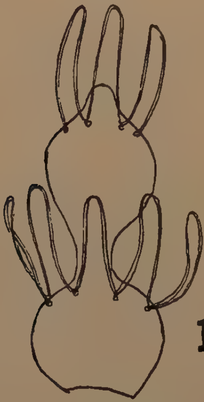
The specimens of this midge were reared by de Nicéville in the Indian Museum from stem galls (Gall No. 268) on *Salix elegans* collected in Tehri-Garhwal, Himalayas (9,000 ft.). The species does not appear to have been collected since then.

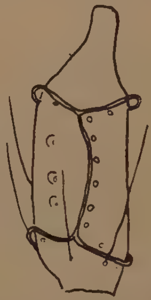
According to Coquillett, the midges are dark brown, with the mesonotum black, antenna about ½ the length of the body in the male and ⅓ the length of the body in the female, with 19 segments. Length 1·5–2·0 mm. de Nicéville (1903) published a figure of the gall.

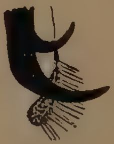
Tribe Asphondyliini.

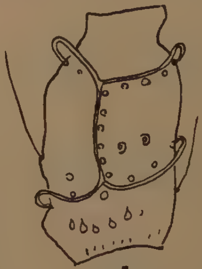
Asphondylia ricini, sp. nov. ✓

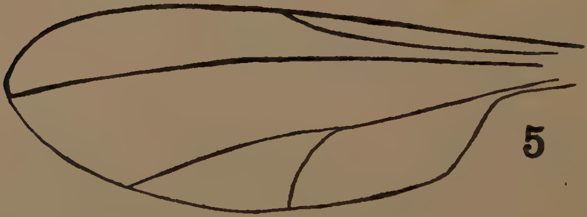
Female.—Length 2·5–3 mm. Dark reddish-brown. Palpi (fig. 7), including the palpiger, triarticulate; segments unequal in length, moderately densely clothed with long, slender setae; first segment cylindrical, stout, short, length twice the thickness; second segment somewhat stouter than the first, also stouter apically than basally, 1½ times the length of the first; third segment the longest, nearly three times the length of second, elongate-oval, rather pronouncedly compressed, basally it is nearly half as thick as the apex of the second segment, relatively more densely setose than the rest of the palpal segments, apically produced into a short, blunt, indistinct nipple-like process. Antenna black, segments 14 ; scape twice as long as thick ; pedicel nearly half the length of the scape, globose, narrower ; third segment longest, its length nearly ten times as long as its diameter which is about half that of the pedicel, somewhat abruptly thicker just at the base than elsewhere, stem short, with a length about $\frac{1}{20}$ and a diameter about $\frac{2}{3}$ that of the segment ; basal whorl of circumfila at the basal half of the segment and apical whorl close to the tip, the two whorls being connected together by an almost straight longitudinal band, closely applied to the surface of the segment ; fourth segment nearly as thick as the third but only $\frac{3}{4}$ of its length, circumfila whorl similar to that of the third ; fifth segment somewhat shorter

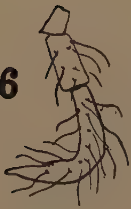


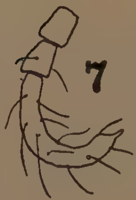












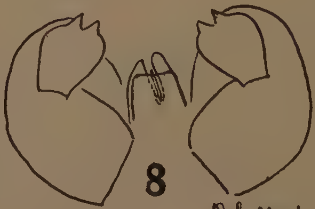


Fig. 1.—*Amradiplosis echinogalliperda*, sp. nov. Fourteenth antennal segment of male.
Fig. 2.—*Amradiplosis echinogalliperda*, sp. nov. Fourteenth antennal segment of female.
Fig. 3.—*Amradiplosis echinogalliperda*, sp. nov. Claw of female.
Fig. 4.—*Amradiplosis echinogalliperda*, sp. nov. Fifth antennal segment of female.
Fig. 5.—*Asphondylia ricini*, sp. nov. Wing of male.
Fig. 6.—*Asphondylia ricini*, sp. nov. Palpus of male.
Fig. 7.—*Asphondylia ricini*, sp. nov. Palpus of female.
Fig. 8.—*Asphondylia ricini*, sp. nov. Genitalia of male.

than fourth and nearly five times its own diameter, circumfila as in the fourth segment, stem about $\frac{1}{3}$ the length of the segment and nearly $\frac{3}{4}$ its thickness ; sixth, seventh and eighth segments equal to the fifth ; ninth segment somewhat shorter and stouter ; tenth about $\frac{2}{3}$ the length of the fourth, somewhat shorter than the ninth ; eleventh about $\frac{2}{3}$ the length of the ninth and a little more than half the fourth, but stouter as the diameter is nearly half the length ; twelfth segment nearly $\frac{5}{8}$ the length of the eleventh but nearly as thick ; thirteenth segment half that of the eleventh and also nearly $1\frac{1}{4}$ its diameter ; fourteenth segment subglobose, in diameter equal to that of the thirteenth segment. Mesonotum black, with the submedian lines moderately clothed with white setae ; scutellum very dark reddish-brown, moderately densely clothed with white setae. Wing as in fig. 5. Claws moderately stout, evenly curved. Empodium very slightly longer than the claws. Acicula of ovipositor when exserted nearly $\frac{1}{4}$ the length of the abdomen ; dorsal pouches moderately large, dark brown.

Male.—Length 2·0–2·5 mm. Black or very dark brown. Palpi (fig. 6) triarticulate, moderately densely clothed with long slender setae, almost similar to that of the female ; first segment nearly twice as long as thick, not so stout as in female ; second segment stouter than the first, subclavate and also nearly twice as long ; third segment like that of the female but slightly more than three times the length of the second. Antenna black, relatively more densely setose, segments 14 ; scape and pedicel nearly as in female ; third segment almost four times as long as thick and as stout as the pedicel, circumfila of the basal whorl almost at the basal fourth of the segment, stem almost obsolete ; fourth segment almost equal to the third ; fifth segment about $\frac{4}{5}$ the length of third, nearly as stout ; sixth segment equal to the fifth ; thirteenth and fourteenth segments equal, each a little shorter than the sixth. Mesonotum and scutellum black. Wing (fig. 5) almost similar to that of the female, though somewhat smaller. Claw as in female, equal to empodium. Genitalia (fig. 8) small, dark brown, moderately setose ; dorsal plate truncated, broadly emarginate ; ventral plate deeply bilobed, lobes oval, longer than the dorsal plate ; basal clasp segment stout ; terminal clasp segment short, obpyriform, black, nearly $\frac{1}{3}$ the size of the basal clasp segment, bidentate apically, teeth short, widely gaping.

Holotype female, *allotype* male dissected on slides ; *paratype* on slide. Received from the State Entomologist, H.E.H. Nizam of Hyderabad, Deccan labelled : " H. M. Farm, 20-x-1945, castor (*Ricinus communis*) capsules, coll. M. K. Alimulla ".

The State Entomologist writes that this species is sporadically a serious pest of the castor-oil plant in Hyderabad.

This species approaches *A. pongamiae*, Felt, and *A. menascéi*, Mani, from both of which it is easily distinguished by the different proportions of the palpal and antennal segments and by the genitalia of the male.

Tribe Itonididini.

Odinadiplosis odinae, Mani.

Odinadiplosis odinae, Mani, 1935. Rec. Indian Mus., **37,** p. 436.

I refer to this species the midges bred from the galls (fig. 9) on petioles and veins of the leaves of *Odina wodier* Roxb. (Gall No. 198), collected by Mr. R. D. Saksena in the jungles of Jhabua in Central India on 19-vi-1945.

The species was originally described from Tanjore and Calcutta and has also been collected from Udaipur in Rajputana (Mani, 1938 & 1943).

Amradiplosis,* gen. nov.

Eyes confluent above. Palpus triarticulate. Antennae with 14 segments, all binodose in the male, the enlargements subglobose, stems nearly as long as the enlargements ; circumfila loops (fig. 11) regular, moderately long, *i.e.* as long as the

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* Amra is the Sanskrit for mango.

enlargements, in two whorls of 8 loops each; in the female segments cylindrical, short, with short stems, circumfila (fig.4) in two short whorls connected by a longitudinal loop; third and fourth segments fused together. Wing (fig. 15) short, broad, hyaline, densely clothed with black setae; third vein reaching costa at apex; fourth vein obsolete; fifth vein forked; no cross-vein. Claws (fig. 3) dentate on all legs. Empodium well developed. Male genitalia (fig. 10) small, sparsely setose; dorsal plate broad, slightly emarginate, broadly and slightly lobed, the lobes broadly rounded; ventral plate much narrower and longer than the dorsal plate, its incision deeper and the lobes almost linear and obtusely rounded apically; harpes broad, much longer than the basal clasp segment which is moderately broad, not lobed; terminal clasp segment slender, a little longer than the basal clasp segment, unidentate. Ovipositor very short and inconspicuous.

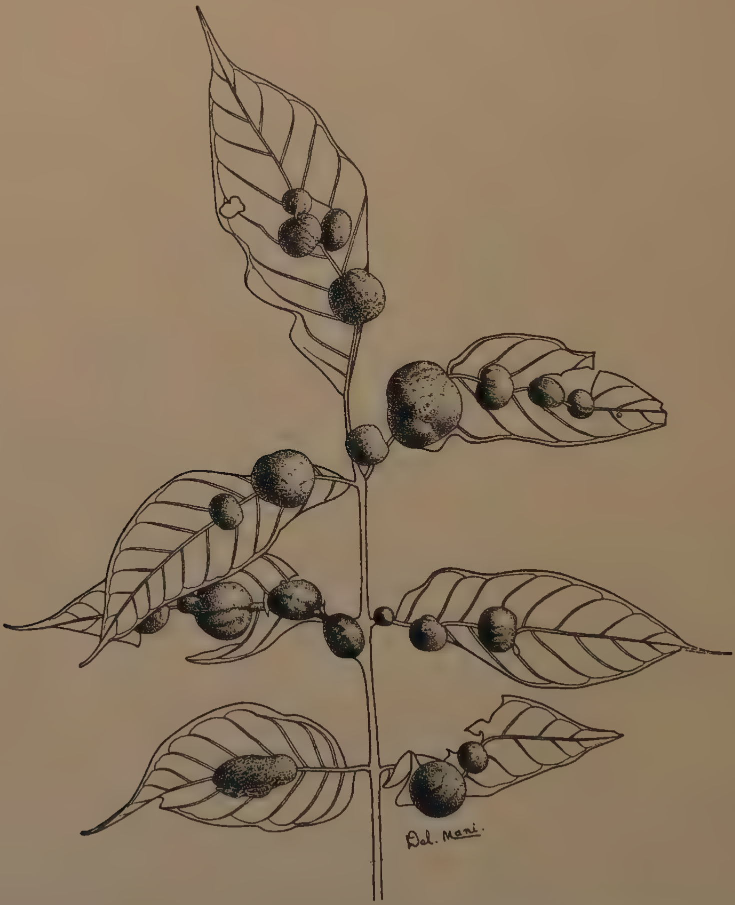


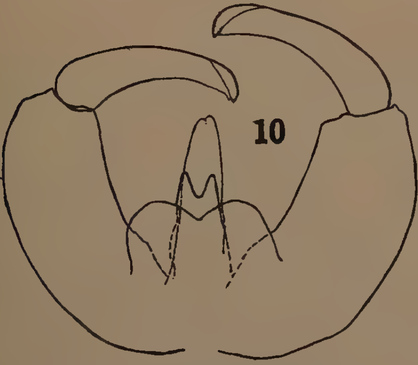
FIG. 9.—Gall No. 198 on leaf of *Odina wodier* Roxb. caused by *Odinadiplosis odinae* Mani.

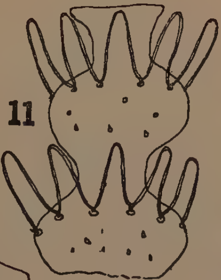
Genotype :—*Amradiplosis echinogalliperda*, sp. nov.

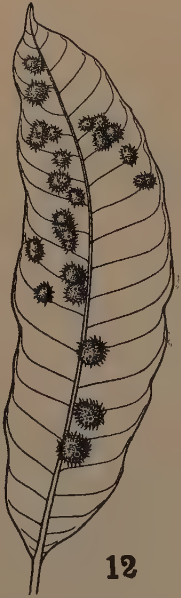
This genus runs to couplet No. 52 in my key (Mani, 1946) to the Oriental genera of the ITONIDIDAE (Mani, 1946) and presents a superficial resemblance to *Mangodiplosis* Tav. (1917). It is readily distinguished from all the genera of the Bifila series of the Itonididini by the triarticulate palpi, unidentate claws, wholly binodose antennal segments of the male and by the male genitalia.

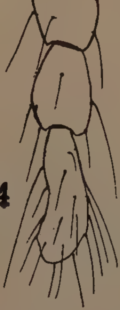
Amradiplosis echinogalliperda, sp. nov.

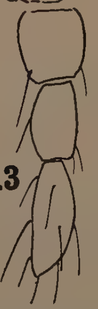
Female.—Length 1·75 mm. Stout, dark red and sparsely setose. Palpi (fig. 14) triarticulate, $\frac{1}{3}$ the height of head, sparsely setose; first segment broadly oval, slightly thicker than long, broader at base, apical diameter nearly $\frac{2}{3}$ the basal; second segment $1\frac{1}{4}$ the length of the first, nearly as thick, stout subapically; third segment longest, a little less than twice the length of the second, subclavate, much more slender than second, only half as thick as the second at base, relatively more densely clothed with long setae. Antenna black, less than $\frac{1}{4}$ the length of body, segments cylindrical, 14; scape very short, transverse, thickness nearly $1\frac{1}{3}$ the length, broadest apically; pedicel subglobose, diameter a little less than the greatest thickness of scape; third segment nearly twice as long as thick, stem short, hardly $\frac{1}{20}$ the length of the segment; fourth segment fused with the third, nearly $\frac{6}{7}$ the length of the fourth, stem distinct, $\frac{1}{6}$ the segment; fifth segment a little less than the fourth, and circumfila as shown (fig. 4); sixth segment equal to fifth; seventh equal to the eighth; ninth, tenth and eleventh segments equal to the eighth; stems of segments seven to eleven prominent, being relatively longer and more slender than those of the basal segments of antenna; twelfth segment a little longer than eleventh, stem a little over $\frac{1}{3}$ the segment; thirteenth segment a little longer than the twelfth and $1\frac{1}{5}$ the length of the fifth; fourteenth segment (fig. 2) somewhat longer than thirteenth, nearly $1\frac{1}{3}$ the length of fifth, apically produced into a subtruncate prolongation about $\frac{1}{4}$ the length of the segment. Mesonotum black. Scutellum reddish. Wing black, short and broad as in fig. 15. Claw (fig. 3) unidentate, moderately stout, evenly curved. Empodium slightly longer than claw. Abdomen half the length of body. Ovipositor short, lobes short, oval.

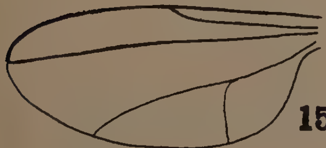












Del. Mani.

FIG. 10.—*Amradiplosis echinogalliperda*, sp. nov. Genitalia of male.
FIG. 11.—*Amradiplosis echinogalliperda*, sp. nov. Fifth antennal segment of male.
FIG. 12.—Gall No. 196 on leaf of *Mangifera indica* Linn. caused by *Amradiplosis echinogalliperda*, sp. nov.
FIG. 13.—*Amradiplosis echinogalliperda*, sp. nov. Palpus of male.
FIG. 14.—*Amradiplosis echinogalliperda*, sp. nov. Palpus of female.
FIG. 15.—*Amradiplosis echinogalliperda*, sp. nov. Wing of female.

Male.—Length 1·5–1·7 mm. Stout, red, and sparsely setose. Palpi (fig. 13) triarticulate, relatively somewhat shorter than in female, first segment globose; second segment slightly longer, broadly oval, somewhat broader subapically than basally; third segment 1⅓ the first segment and 1⅕ the second, oval, subapically armed with short, stiff, erect setae; on the left side of the allotype the third palpal segment is nearly 1½ times the length of the third segment of the right palpus and almost as in female. Antenna long, pale brown; segments 14, all binodose; circumfila in two whorls of twelve loops each, with the loops as long as the stems or slightly longer; scape short, widest at apex, width to length in the ratio of 2 : 1; pedicel transverse, about ⅔ the width of the scape, and in length half the width, widest at base; third and fourth segments fused together; basal enlargement of fourth segment with a diameter nearly 1½ times its length, basal stem half the length of the basal enlargement, apical enlargement equal to the basal, apical stem more slender than basal stem and nearly 1½ times the length; fifth segment (fig. 11) almost equal to the fourth, enlargements, stems and circumfila as in fig. 11; sixth segment equal to fifth but distinctly more slender, apical enlargement with a diameter a little less than that of basal enlargement; seventh, eighth and ninth segments equal to fifth but gradually growing more slender, with the stems also gradually becoming longer and slender; tenth segment with apical stem slightly longer than apical enlargement and nearly twice as long as its own diameter; eleventh segment like the tenth; thirteenth segment with the apical stem nearly three times as long as thick; fourteenth segment about ⅚ the length of the penultimate segment, apical enlargement pyriform and produced into a short, broad, rounded and conical tip as in fig. 1. Mesonotum black. Wing black, short and broad as in female. Claws unidentate as in fig. 3, stout, evenly curved. Empodium longer than claws. Genitalia (fig. 10) slender, sparsely setose, brown; dorsal plate broad, bilobed by a slight incision in the median line, the lobes broadly rounded, slightly emarginate; ventral plate longer than dorsal plate, more heavily chitinised, narrow, bilobed by a relatively deeper median apical incision, the lobes narrow, obtuse apically; harpes broad, stout, very much longer than the ventral plate and nearly as long as basal clasp segment; basal clasp segment slender, simple, nearly twice as long as wide; apical clasp segment slender, slightly curved, relatively more heavily chitinised, black, nearly ¾ the length of the basal clasp segment, thickness at base about ¼ its length, apically nearly half as thick as at base, unidentate.

Holotype female, *allotype* male dissected on slides; *paratypes* females in spirit. 12 females and 1 male reared at Agra between 12–18-x-1945 from echinate gall No. 196 on leaf of *Mangifera indica* Linn., coll. M. S. Mani, New Forest, Forest Research Institute, Dehra Dun, U.P., 7-x-1945.

I collected the gall of this species first in December 1934 at Calcutta and later in March 1941 at Dehra Dun but the midges could not be reared on either occasion (Mani, 1935).

Galls epiphyllous, regular, globose, usually free, sessile, cystiferous, very much resembling a miniature sea-urchin (fig. 12), about 5–8 mm. in diameter, up to about fifty galls per leaf but generally only about twenty; the spinous processes are fleshy, parenchymatous and irregularly elongate-conical; green when young but turning reddish when ripening and finally almost black.

The pupation of the midge takes place within the gall and lasts about eight days during October.

Orseoliella, Kieffer.

Orseolia (partim) Kieffer & Massalongo, 1909. Marcellia, **7,** p. 125.
Orseoliella, Kieffer, 1912. Marcellia, **11,** p. 231.
Orseoliella, Mani, 1934. Rec. Indian Mus., **36,** p. 432.

Kieffer (1912) described the flagellate antennal segments of the male of the genotype, *O. javanica* (Kieff. & van Leeuw.), as having five transverse bands of circumfila—figured by Mani, 1946—and later (1913) included this as one of the diagnostic characters of the genus *Orseoliella*. He has also described the empodium as being a little longer than the claws in the genus.

In 1921, Felt identified the midges reared from galls on *Imperata arundinacea* (=*cylindrica*) from India as belonging to *O. javanica* (Kieff. & van Leeuw.) and remarked as follows : " There is little question concerning the identity of this insect with the species originally described from Java, despite the fact that the material at hand shows no such peculiar arrangement of circumfila as depicted for the female antennal segments in the original description, and later elaborated in greater detail by Kieffer in Fascicle 152 of Genera Insectorum. We are inclined to believe that the artist was misled at the outset in regard to the arrangement of these peculiar structures." Further, the circumfila of all the other species so far known from India are normal, *i.e.* have only an apical and a basal transverse band in the female. The empodium moreover is subject to a great deal of variation even within the limits of the same genus and is therefore unreliable as a generic character.

Altogether five species of *Orseoliella* are so far known from India, including one new species described here :—*O. javanica* (Kieff. & van Leeuwen-Reijnvaan), *O. apludae*, Felt, *O. graminis*, Felt, *O. bengalensis*, Mani and *O. ischaemi*, sp. nov. They may be separated as follows :—

Key to Species.

1. Males 2
   Females 5
2. Empodium shorter than claw 3
   Empodium longer than claw 4
3. Antenna longer than body ; ventral plate equal to basal clasp segment .... *javanica* (Kieff. & van Leeuw.)
   Antenna $\frac{2}{3}$ the length of body ; ventral plate $\frac{3}{4}$ the length of basal clasp segment *ischaemi*, **sp. nov.**
4. Ventral plate half the length of basal clasp segment .... *bengalensis*, Mani
5. Empodium as long as claw 6
   Empodium a little longer than claw 7
6. Antenna half the length of body .... *ischaemi*, **sp. nov.**
   Antenna equal to body .... *graminis*, Felt
7. Antenna $\frac{3}{4}$ the body .... *apludae*, Felt
   Antenna equal to the body .... *javanica* (Kieff. & van Leeuw.)

Orseoliella ischaemi, sp. nov.

Male.—Length 3 mm. Reddish-orange. Densely setose. Palpi quadriarticulate, short, sparsely setose, almost as in female. Antenna $\frac{2}{3}$ length of body, orange ; segments 14, all binodose, basal enlargements subglobose or globose, apical enlargements subcylindrical, stems long ; circumfila in three whorls, each of short, regular loops, first whorl at apical $\frac{1}{3}$ of the basal enlargement, second whorl subbasally and the third whorl subapically on the apical enlargement ; scape obconical, nearly as long as wide at apex ; pedicel subglobose, more slender than scape ; third segment with the basal enlargement somewhat oval and in diameter equal to that of pedicel, length nearly $1\frac{1}{5}$ its diameter, basal stem with a length $\frac{1}{2}$ the diameter of the basal enlargement and its thickness equal to its length ; apical enlargement twice as long as thick, $1\frac{1}{2}$ the length of the basal enlargement, stem somewhat more slender than that of the basal enlargement but nearly equal in length; fourth segment, equal in length to the third, basal enlargement about $\frac{1}{6}$ the length of the segment, globose, basal stem with a length about $\frac{2}{3}$ the diameter of the basal enlargement and thickness about $\frac{4}{5}$ its length, length of apical enlargement $\frac{3}{7}$ that of the segment and twice the diameter of the basal enlargement, thicker subapically, slightly constricted at basal third, stem slightly longer and more slender than the basal stem, its diameter about $\frac{2}{3}$ its length; fifth segment as in fig. 19, equal to the fourth, basal enlargement globose, length of basal stem a little over $\frac{1}{2}$ the diameter of the enlargement and $1\frac{1}{5}$ its own diameter, somewhat more slender than the basal stem of the fourth segment, apical enlargement and apical stem similar to those of the preceding segment but in the latter case somewhat more slender; sixth and seventh segments like the fifth but more slender, stems relatively longer and more slender; eighth segment (fig. 18) slightly shorter, apical enlargement distinctly binodose so that the segment itself is trinodose, the constriction being at the basal third as in fig. 18, apical stem half the length of apical enlargement and $2\frac{1}{2}$ times its own diameter; ninth segment onwards gradually growing more and more slender, though nearly equal in length, the stems also progressively longer and more slender; fourteenth segment reduced apically to a slender, truncated tip. Mesonotum black. Scutellum brown. Wing long, moderately narrow, nearly twice as long as broad. Legs long, orange. Claws simple, slender, slightly curved. Empodium narrow, slightly longer than claws. Abdomen nearly $\frac{2}{3}$ the length of body, slender, densely setose, orange-brown. Genitalia large, densely setose, deep brown; basal clasp segment large, with a large obtuse lobe basally (fig. 20), apical half slender, narrow, nearly as wide as the apical clasp segment; terminal clasp segment moderately long, slender, slightly curved, about $\frac{2}{3}$ the length of the basal clasp segment, conspicuously rounded and minutely and bluntly bidentate at tip; dorsal plate damaged; ventral plate large, simple, slightly emarginate, reaching nearly up to $\frac{3}{4}$ the basal clasp segment; style stout, a little longer than the ventral plate.

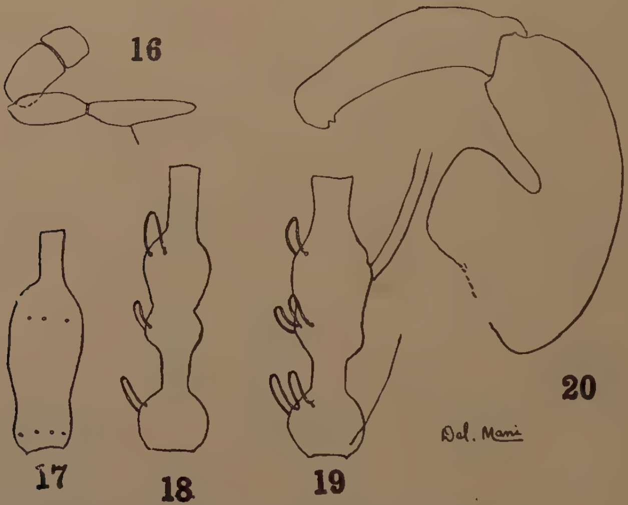


FIG. 16.—*Orseoliella ischaemi*, sp. nov. Palpus of female.
FIG. 17.—*Orseoliella ischaemi*, sp. nov. Eighth antennal segment of female.
FIG. 18.—*Orseoliella ischaemi*, sp. nov. Eighth antennal segment of male.
FIG. 19.—*Orseoliella ischaemi*, sp. nov. Fifth antennal segment of male.
FIG. 20.—*Orseoliella ischaemi*, sp. nov. Genitalia of male.

Female.—Length 4 mm. Red. Palpus (fig. 16) short, slender, sparsely setose, quadriarticulate ; first segment short, quadrate, 1½ times as long as thick ; second segment stout, twice the length of the first ; third segment slightly longer, somewhat abruptly reduced apically ; fourth segment slender, shape as in fig. 16, reduced beyond basal third, 1⅔ the third segment, subtruncate at tip. Antenna half the length of body, brown, segments 14, subcylindrical, moderately densely setose ; pedicel broadly oval, nearly 1¼ as wide as long ; third segment nearly as long as that of the male, five times as long as thick, somewhat stouter subapically, stem ⅐ the segment, its diameter a little over half the length ; fifth segment a little over ¾ the length of the third, rather stout subapically, stem relatively more slender, about ⅓ the length of the segment and in diameter about ⅖ its length ; eighth segment (fig. 17) a little shorter than fifth segment, stouter subapically than basally, stem ¼ the length of segment, its diameter a little less than half its length ; tenth segment nearly ¾ the length of the fifth, stem about ¼ its length ; fourteenth segment with a terminal pyriform process constricted off conspicuously from the rest and nearly half the length of the segment. Mesonotum black. Scutellum dark brown. Wing nearly as long as body and also twice as long as broad. Legs reddish-brown. Claws simple, slender, evenly curved. Empodium equal to claws. Abdomen red, a little more than ¾ the body. Ovipositor short, exserted, lobes short, narrow, spatulate-oval, nearly thrice as long as broad.

Holotype male, *allotype* female on a slide. Coll. Y. R. Rao, Coimbatore, South India, from galls on *Ischaemum ciliare* ; 31.viii.1916.

Parallelodiplosis javanica, Felt.

Parallelodiplosis javanica, Felt, 1921. Treubia, **2**, pp. 89–92.

I refer to this species the midges bred from stem gall on *Panicum indicum* at Calcutta. Coll. Mani, July 1934.

The species was originally described from Java on the same plant and this is the first record of the species from India.

Asphondylia ricini, Mani.

Mr. A. S. Rao, Entomological Assistant, Oil seeds Research Scheme, Hyderabad, Deccan, sends me the following note on this species :—

During the course of the survey of the pests of the castor-oil plant in Hyderabad State a Cecidomyiid was found attacking the capsules of the plant. As far as I am aware this is the first time a midge pest is observed on this plant. Both the capsules and the male flowers are attacked by the larvae. The capsules do not develop, become deformed into a gall and finally drop off prematurely. In each deformed capsule usually four to five maggots of the midge occur. The maggots are white. Staminate flowers do not open as a result of the attack by the maggots, causing non-pollination of even the normal pistillate flowers which have escaped the attack. Oviposition presumably takes place inside the tender buds, the eggs not having been observed so far in the field. The full-grown maggot becomes yellow and pupates within the damaged plant parts. The pupae are reddish-brown and measure 3 mm. long.

As far as the present observation goes, the percentage of incidence of the attack is rather high and severe damage is caused to the tender parts. This leads to extensive failure of crop.

A species of *Bracon* and another of *Eurytoma* are parasitic on the larvae of the midge ; the *Eurytoma* is perhaps a hyperparasite.

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THE EFFECTS OF HOUSE SPRAYING WITH PYRETHRUM AND WITH DDT ON *ANOPHELES GAMBIAE* AND *A. MELAS* IN WEST AFRICA.

By R. C. Muirhead Thomson, D.Sc.,
Colonial Medical Research.

L.C.

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The observations described in this paper were made during the latter part of an 18-month tour of research in Lagos, Nigeria. The first part of the tour was mainly devoted to a study of the house-haunting habits of *Anopheles gambiae*, Giles, and *A. melas*, Theo., in relation to feeding and resting. During the course of that work a great deal of information about the normal habits of these two Anophelines was gained, particularly with regard to the use of outside resting places, and the times of entering and leaving habitations.

In addition to this a great deal of local knowledge accumulated, so that when the time came to experiment on house-spraying there were three ideal localities available. One isolated group of villages in which a steady supply of both *gambiae* and *melas* was assured; another small village with particularly suitable outside resting places for mosquitos; and thirdly, an isolated group of huts where the numbers of mosquitos remained sufficiently high to make observations on single houses worth while. Field workers in the tropics will appreciate how unusual it is to find such an ideal combination of conditions in villages only a few miles apart.

Before going on to describe the house spraying experiments, there are some facts about the normal behaviour of mosquitos that came to light in the first part of the tour, and that have a direct bearing on later experiments. A detailed account of the behaviour of *gambiae* and *melas* in houses will be given in a separate paper to be published shortly, and the following is a brief summary of the relevant parts of that paper.

House-haunting Habits of *A. melas* and *A. gambiae*.

A. gambiae and *A. melas* are well known to be domestic in habit, in that the adults can commonly be found resting by day inside dark houses, and bedrooms in particular. Both in Sierra Leone and Nigeria both species have been found to have normally a 48-hour cycle to digest the blood meal and develop the ovaries. For example, females feeding on say Monday night will be ready to lay eggs on Wednesday night, and are capable of taking another blood meal on the night on which oviposition takes place.

The females resting in houses by day can be divided into "blood-feds" and "gravids", the former will have fed on the previous night, and the latter on the night before that. If these mosquitos were completely domestic in habit, resting nowhere else but in human habitations, the number of blood-feds and gravids would be expected to be roughly equal. But the house catches always showed a much greater preponderance of blood-fed over gravid, showing that the gravids at least must make use of other resting places as well.

The knowledge that there must be day-time resting in places other than habitations encouraged the search for outside resting places in the face of continued negative results, until finally resting adults were discovered in holes and crevices of well shaded termite mounds, in folds and fissures on the shaded trunks of large forest trees and in certain heavily shaded earth banks. Only in the last place could the finding of adults regularly be relied upon.

If the unfed and unfertilised females found in these places are ignored, not only are the gravids to be expected found, but also an almost equal number of blood-fed females. If the illustration used above is adopted it can be said that, of the adults feeding in human habitations on Monday night, many will remain in the house on Tuesday as blood-feds, but others will leave the house before daybreak and rest outside. Some of the blood-feds remaining in the house on Tuesday will stay on in the house and be found there as gravids on the following day, Wednesday, finally leaving to lay eggs on Wednesday night : but other blood-feds leave the house on Tuesday night, and are found as gravids in outside resting places on Wednesday.

In order to find out when, and to what extent, mosquitos leave the house during the 48-hour gonotrophic cycle, it was possible to show first of all that mosquitos leaving the house between dusk and dawn are attracted by the faint light coming in through windows, open doors, wide spaces in the eaves, and so on. So strong is this attraction that it can be used to form a most efficient trapping system for *Anopheles* leaving the house during the hours of darkness. (During the bright light of day mosquitos that have not left the house by dawn tend to remain inside the house, and are not attracted by daylight coming in at the windows.)

Small huts were constructed with mud walls and palm thatch roofs, in such a way that while hungry mosquitos had no difficulty in entering under the eaves, the only obvious way out of the hut was through a small window over which a one-way trap was fixed. From inside the hut, between dusk and dawn, the only light coming in was through the 1-foot square window opening ; the door being covered on the inside with black curtain, and all small chinks of light eliminated. Further details about the trap will be given in the fuller publication, but for the present purpose it is sufficient to state that by means of this window trap the conclusions reached above were confirmed and extended, and it was shown that, even when the hut offered ideal dark day-time resting places, many blood-fed mosquitos leave the house at dawn after feeding and that of those remaining in the house a large proportion leave at dusk the following night.

In the average village house there is much more light coming through open doors, wide spaces between walls and eaves, windows, holes in walls, etc. ; the number of mosquitos caught in such houses during the day, although high, may represent only a small proportion of the mosquitos actually feeding there at night.

Another point of importance is the feeding activity of females in the houses. With *gambiae* and *melas* it was found that there is very little feeding before 10 p.m. and that 90 per cent. of the blood feeding takes place after midnight. From midnight onwards there is a steady increase in feeding activity, reaching a peak in the hour or two before dawn. When it is remembered that many blood-fed females leave the house at dawn, it is clear that many of them must do so within a short time of feeding, and that it is possible for the mosquito to spend only an hour or two of the 48-hour gonotrophic cycle actually in the house.

Having established a simple but effective method of trapping mosquitos leaving the house in as natural a way as possible, it was but a step to find out how the behaviour was affected by spraying the house with insecticide. The preliminary work, however, has already shown that *melas* and *gambiae* are not so highly domestic as had been imagined, and that their habits do not lend themselves quite so well as was thought to direct attack against the adults.

Effect of spraying Houses with Pyrethrum in Kerosene.

The spray-killing of adult mosquitos in houses is now known to be an effective anti-malaria measure in many parts of the world ; the most conclusive results coming from South Africa and India. A general review of the whole subject has been given recently by Senior White, Ghosh & Venkat Rao (1945) and some of the points raised will be discussed in the course of this paper.

In West Africa there has been an immense amount of spray-killing with pyrethrum in kerosene in houses during the war years, but all claims to its efficacy have been based purely on analogy with findings in other countries, and there is really nothing conclusive to show whether or not its use has been justified. One of the few published reports (Eddey, 1944) has been concerned principally with labour or costs, the success of the method being assumed on the principle that if spray-killing is effective against *A. gambiae* in South Africa, it must also produce results in West Africa.

The crucial experiment of finding how spray-killing alone affects village malaria over a period of years—as done by Russell and Knipe (1939–41) on *A. culicifacies*, Giles, in South India—has yet to be done in West Africa. In the present paper, however, although only the entomological aspect of the problem has been dealt with, the results are sufficiently discouraging to render further large scale experiments unlikely.

Experiment 1. Mass spraying of villages.—The object of this first experiment was to find out what effect mass spraying of all rooms in an isolated group of villages had on the house population of *A. melas* and *A. gambiae*, and on the infectivity of the mosquitos.

In an experiment of this kind it is usual and indeed desirable to have some sort of control, either in the form of untreated rooms or untreated villages. In the present experiment it was considered that leaving untreated rooms in the sprayed villages might introduce complications ; there was also at that time no suitable village where conditions were sufficiently similar to those of the sprayed villages to justify its use as control. The experiment was done at a time of the year when there was a very steady production of *A. gambiae* and *A. melas* in this particular group of villages and, as comparison therefore, conditions during the month before spraying started were used, and conditions after spraying.

The experiment was started in December 1945 in a group of villages between Ebute Metta and Apapa, near Lagos. Collections were made twice a week for a month in the villages of Alagbede, Senu and Iganmu by hand-catching in bedrooms, the number searched varying from 12 to 27 at a time. The day catch remained remarkably steady throughout the preliminary survey, the numbers of females per 10 bedrooms varying from 26 to 38, with a mean of 33. Sixty per cent. of the adults identified by egg characters were *melas* and 40 per cent. were *gambiae*. The sporozoite rate of 485 mosquitos dissected was 3·5 per cent.

When the preliminary survey was complete, spraying of the village houses was started. The particular brand of pyrethrum extract used was " Pyefly " diluted 1 in 59 parts of kerosene, and sprayed at the rate of ½ to ¾ ounce per room of 1,000 cubic feet using " Maney " continuous hand sprayers. The houses were well adapted for spraying ; tests with sheets spread on the floor showed that spraying was exceedingly effective in bringing down all the *Anopheles*, and that among those knocked down recovery was exceedingly rare.

Every precaution was taken to eliminate the chances of infected mosquitos coming in from uncontrolled areas. In addition to the villages named above two more, Ajegunle and Abulenla, were included in the spraying programme—bringing the total rooms sprayed to 150—and collections were restricted to the same rooms as before, which now formed the centre of a half-mile radius within which nearly all houses were being sprayed. The number of locked rooms in these villages remained between 5 and 10 per cent., but they were all sprayed through doors or windows, or over the walls of adjacent rooms.

Senior White and Venkat Rao (1944) have pointed out the importance of adjusting the spraying programme to suit the gonotrophic cycle of the mosquito. With efficient vectors like *A. melas* and *A. gambiae*, which have a 48-hour cycle, it was not considered worth attempting anything less than 4 applications a week, the days selected being Monday, Tuesday, Thursday and Friday, which on Senior White's convention would be represented as $\underset{\overline{1\ \ 2\ \ 1}}{2\text{–}2\text{–}2}$. If the mosquito used the house as a resting place every other day, then spraying on this spacing, would give 2 to 3 chances of it being caught by the spray before it could become infected (which at the earliest would happen when it was 9 days old).

Two weeks after regular spraying had started, routine hand collections were resumed in the same houses used in the preliminary survey, and continued for 5 weeks. The number of mosquitos now caught was reduced to a third of its former figure, with a mean catch of 11 females per 10 bedrooms. Of 400 mosquitos dissected, the sporozoite rate was 3 per cent., infected mosquitos still being found at the end of the experiment.

The results are shown in Table I.

TABLE I.

Mass spraying of village houses with pyrethrum in kerosene. Incidence and infectivity of *melas* and *gambiae*. December 1945–April 1946.

Treatment	Number and period of collections	*Gambiae* and *melas* per 10 bedrooms	Mosquitos dissected	Glands positive	Sporozoite rate
Preliminary survey. Houses untreated ...	8 18/xii to 19/i	33	485	17	3·5%
All houses sprayed four times weekly from 19/i	15 1/ii to 7/iii	11	400	12	3·0%
All houses sprayed six times weekly from 7/iii	7 14/iii to 2/iv	12	247	18	7·3%

Various observations in these villages have shown that at the mosquito densities encountered at this time of the year, ½ mile is about the maximum flight range. Mosquitos may be extremely scarce in villages less than ½ mile from large foci of mosquito production ; two villages in the experimental area show at times completely different populations, one being almost entirely *melas* and the other *gambiae*, although the villages are only half a mile from each other. With the nearest uncontrolled source of infected *gambiae* and *melas* 1½ miles from the sprayed villages, it was reasonably certain that the activities of the mosquitos were mainly confined to the group of sprayed villages.

The reduction in numbers of *melas* and *gambiae* in houses following spraying was much less marked than Senior White found with *A. fluviatilis*, James, in India ;

but the general similarity of the results, with the sporozoite rate unaffected, is so striking that there seems to be some common principle involved.

The spraying was now stepped up to 6 days a week, and observation continued. *Gambiae* and *melas* had been about equally abundant in village houses up to this time ; but now, in March, *gambiae* was becoming more dominant, forming 80 per cent. of the catch. As a result neither the densities of mosquitos in houses, nor their infectivities, can be compared directly with those of the previous spraying experiments, but the results—in Table I—show clearly that even this intensive spraying was unable to bring about a substantial reduction, either in the number of mosquitos in the houses, or in the sporozoite rate.

Experiment 2. Observations in experimental huts.—In the early part of this paper the simple method of trapping mosquitos leaving the hut by means of a window cage or trap was described. These experimental huts were constructed of mud walls and palm-thatched roof after the fashion of ordinary village houses in this area ; and it is reasonably certain that the behaviour of mosquitos in these huts did not differ fundamentally from that in the nearby African villages.

In the experiment on mass spraying of villages described above, it was established that the day catch of mosquitos in a sprayed house was only about one-third of what it was before treatment started. But from previous knowledge of the movements of *gambiae* and *melas* in and out of houses, it is known that this fall in the house catch cannot be taken as a sure indication that there is a similar fall in the whole existing population of mosquitos inside and outside of the village houses. The following observations in the experimental huts are intended, therefore, to show exactly what happens to the mosquitos in a house that is being sprayed regularly with pyrethrum in kerosene.

The huts were those previously used in behaviour studies. Two boys slept in each hut, and the huts were provided with window traps to catch all mosquitos leaving the house. These huts were designed in such a way that the low roof was within easy reach of the collectors ; but it was necessary to take certain precautions when collecting resting mosquitos by hand catching. When a first careful search had been completed, the walls and ceiling were lightly brushed with a bunch of grass or twigs, and a second collection made after five or ten minutes. A good many mosquitos escaped the first catch, but very few remained undetected after the double catch.

Experiment 2a.—Two huts were used here. Preliminary catches showed that the number of mosquitos feeding in the two huts occupied by boys was much the same, and that the mean proportion (mostly *melas*) leaving the huts after feeding was about 25 per cent.

One hut was left untreated, while the other was well sprayed *once* with pyrethrum in kerosene. Collections in both huts and their window traps were made on successive days.

The results of 4 series of experiments are shown in Table II, and it is seen that for three days after spraying the number of blood-fed *melas* found in the sprayed hut was much less than in the untreated hut. By the 5th day the effects of the spraying had almost disappeared. On the first and second days after spraying, about ⅔ of the mosquitos which do feed in the sprayed hut, leave after feeding, and are trapped in the window cage. By the third day more blood-fed mosquitos are found resting in the hut than in the window cage.

Under the conditions of this experiment, when the mosquito is offered a choice of feeding in sprayed and unsprayed huts, the pyrethrum in kerosene evidently exerts a repellent effect, not only towards many of the hungry unfed females, but it also, on the first two days after spraying, tends to drive out of the house most of the mosquitos which do manage to feed there.

Experiment 2b.—In the experiments described above the mosquitos were offered the choice of feeding in an untreated hut or in one which had been sprayed once with pyrethrum in kerosene. Under those conditions the feeding in the sprayed hut was much less for three days after spraying than it was in the untreated hut.

The present experiment was designed to see the effect of spraying *all* the experimental huts occupied by man *every* day, and to duplicate more closely conditions in villages which were being mass sprayed.

TABLE II.

Effect of single spraying of experimental hut with pyrethrum-kerosene on successive days catches of blood-fed *melas*, compared with adjacent untreated hut. Four series of experiments.

	Untreated hut			Sprayed hut		
	Window trap	Residue in hut	Total	Window trap	Residue in hut	Total
Before treatment	27	77	104	25	91	116
1st day after spraying	22	39	61	15	5	20
2nd day ,, ,, ...	37	55	92	8	0	8
3rd day ,, ,, ...	48	85	133	24	31	55
4th day ,, ,, ...	55	84	139	33	53	86
5th day ,, ,, ...	25	58	83	12	47	59

In the little isolated group of 6 experimental huts, the three occupied by boys were sprayed each day with pyrethrum in kerosene, while the other three huts were occupied by animals and were untreated. Before the experiment began the attraction of *melas* to the man huts was almost complete, only a very small proportion

TABLE III.

Daily spraying of man huts : animal huts untreated. Effect of daily spraying with pyrethrum-kerosene on biting activities of *melas* as shown by blood-fed females caught.

(*a*) 4 men (2 huts) and 8 animals (3 huts)

	Number of collections	Man huts		Animal huts	
		Window trap	Hut catch	Window trap	Hut catch
Before spraying ...	5	37	199	17	10
Daily spraying of man huts	2	30	4	0	0

(*b*) 6 men (3 huts) and 8 animals (3 huts)

	Number of collections	Man huts		Animal huts	
		Window trap	Hut catch	Window trap	Hut catch
Before spraying ...	2	80	311	1	8
Daily spraying of man huts	3	237	74	2	0

feeding on the animals. The animal aspect of the experiment will be described later; the main object of the experiment was to find out what happens when the mosquitos have no alternative choice of feeding in unsprayed man huts.

Two series of experiments were carried out, using 4 boys (2 huts) in the first, and 6 boys (3 huts) in the second. The number of animals remained constant—3 pigs, 1 calf, and 4 goats.

The results are shown in Table III.

The results of all these experiments show that when all the man huts are sprayed every day, the total number of *melas* feeding in these huts is not greatly reduced, but that there is a very great increase in the proportion of mosquitos that leave the house after feeding.

In the first series (4 boys) before spraying started, 16 per cent. of the blood-fed mosquitos left the house after feeding; after daily spraying started, 88 per cent. left the house after feeding.

In the second, and more conclusive series (6 boys), before spraying started, 20 per cent. of the blood-fed mosquitos left the house after feeding; with daily spraying this figure increased to 76 per cent.

Another way of putting it is to say that in an untreated hut of this kind the number of blood-fed females found resting by day represents $\frac{2}{3}$ to $\frac{3}{4}$ of the total numbers feeding there the previous night; but in the hut sprayed daily with pyrethrum in kerosene the number found in the hut by day represents only about $\frac{1}{5}$ of the total feeding there at night.

The next point that emerges from the experiments is that daily spraying of man huts does not increase the number of *melas* feeding on animals in untreated huts. Experiment 2a shows that the pyrethrum in kerosene has an irritant effect on many blood-fed mosquitos, driving them out of the house after feeding; and also that a repellent effect prevents many mosquitos from entering the house to feed. Experiment 2b suggests that the repellent effect on hungry mosquitos may only be effective when the mosquito is offered a choice of feeding on man nearby in an untreated hut. When all man huts have been treated with pyrethrum it appears as if few of the hungry mosquitos attracted to the man huts are prevented from feeding, otherwise an increase in the number feeding on animals in untreated huts would be expected.

These experiments with huts and window traps also throw some light on the observations of other workers. Ribbands (1946) has suggested that daily spraying of huts at the rate of 25 cc. per 1,000 cubic feet deterred 90 per cent. of *A. minimus*, Theo., from entry on the following night. But it is clear from the observations above that reduction in the daytime catch following spraying does not necessarily mean a corresponding reduction in feeding activity in the hut; and that unless due account is taken of those mosquitos which leave the house shortly after feeding it is almost impossible to say what proportion are deterred from feeding. 34 199

From the present observations, too, it appears as if the mosquitos' reaction to pyrethrum treated huts may vary according to whether an untreated house with suitable host—man—is or is not available. If the mosquito is offered such a choice, then the pyrethrum (or the kerosene or both) does deter many mosquitos from entering the treated hut. If no such choice is available, the feeding activity in treated houses is not greatly reduced.

The observations also show that however efficient spray killing may be in estimating the total house or room catch, it should not be used in the same house more than once a week, otherwise the results may be affected by a shift of mosquitos from inside the house to outside resting places. It also goes without saying that in studying the normal habits of mosquitos in houses, the very last thing one wants to do is to upset the results by using spray killing too frequently.

Discussion on House-spraying with Pyrethrum.

If the findings in the above experiments are applied to the observations made in mass spraying of villages, some explanation of the results can be offered. It is possible that the direct lethal effect of frequent spraying with pyrethrum in kerosene in houses is largely counteracted by the fact that the treated rooms become increasingly unattractive, not to hungry mosquitos, but to mosquitos which have fed in these rooms ; and that more and more of these blood-fed mosquitos leave the house at or before dawn, thus escaping further esposure to day time spraying. When the spraying is done at intervals of one week—a spacing sufficient only for mild vectors like *A. culicifacies*—the repellent effect, or rather irritant effect, has time to wear off between sprayings ; but when spraying is carried out two or more times a week, the irritant effect on the blood-fed mosquitos becomes increasingly important, and the proportion of the population directly affected by the spray becomes smaller and smaller.

The fall in the number of mosquitos resting by day in houses after spraying is now seen to be caused, not by any great reduction in the population as a whole, but rather to a shift from indoor to outside resting places. The number of mosquitos killed by even the most intensive spraying represents such a small proportion of the whole that the average length of life may be little affected, the sporozoite rate remaining at much the same level as before.

DDT as a Residual Spray in Houses.

In the observations on pyrethrum it became increasingly clear that a sound knowledge of the normal behaviour of mosquitos in houses was necessary before the results of house spraying experiments could be interpreted correctly. This is doubly true of DDT, and there is little doubt that not all workers with this insecticide have appreciated how very wide the gap between laboratory and field conditions may be.

The relevant literature will not be discussed in detail till later in this report, but it can be said that the general findings from most countries are that houses treated with DDT remain lethal to mosquitos for periods of one or two or even more months.

In West Africa the mass treatment of village houses with DDT has occupied a prominent place in anti-malaria programmes for the last two years—particularly with the Army control units. There is much information still unpublished, but the general opinion is voiced by Kartman and da Silveira (1946) who say that " The efficacy of DDT residues as a control of *A. gambiae* and *A. funestus* in native villages near Dakar has already been demonstrated by the work of the Army Malaria Survey Detachment." Wilson (1946) considers that in all probability spraying houses with DDT twice a year will be effective.

From these and other observations it is evident that treatment of houses with DDT may reduce the house catch to zero, and keep the figures very low for several months after treatment. This dramatic fall in the house catch has been generally accepted as sufficient evidence of mosquito reduction. It would perhaps be as well at this stage to say that the findings in the present report have revealed unforeseen snags which may limit the value of DDT (in its present form) in kerosene as a residual insecticide in West African village houses, and have also disclosed fallacies in the commonly accepted interpretation of experiments with this insecticide.

In order to obviate any possibility of these results being vitiated by poor insecticidal qualities, samples of the DDT used were brought back to England for testing. I am grateful to Dr. R. A. E. Galley of the Agricultural Research Council for reporting on the samples, the para para content of which was found to be 67 per cent.

Experiment 1. Mass spraying of houses with DDT in Kerosene. Effect on mixed population of gambiae *and* melas.—The villages used were the same as those in which the pyrethrum-kerosene experiments were carried out. The walls and ceilings in 5 villages (about 150 rooms) were sprayed at the rate of 100–125 mg. DDT per sq. ft. using 5 per cent. DDT in kerosene (8 oz. per gallon) at the rate of 1 quart per room 10×10×10 ft. (500 sq. ft. of surface), applied with a 4-Oaks, Kent, pressure-operated knapsack sprayer.

Most of the houses were mud walled, with thatch or corrugated iron roof. Particular care was taken to spray all possible resting surfaces, but hanging clothes, curtains, wall pictures etc., could not be treated. These were, however, of less importance than ceiling and rafters which received thorough treatment.

The fluctuations in mosquito house population were followed as before by hand catching. The results are shown in Table IV.

TABLE IV.

Mass treatment of village houses at rate of approximately 100–125 mg. DDT per sq. ft. using 5 per cent. DDT in kerosene. Villages sprayed: Iganmu, Senu, Alagbede, Ajegunle, and Abulenla (150 rooms).

	Week before spraying	After spraying								
		1 Week	2 Week	3 Week	4 Week	5 Week	6 Week	7 Week	8 Week	9 Week
Rooms searched ...	39	40	39	28	20	39	41	19	19	19
Anopheles found. *Melas* and *gambiae*	104	1	0	0	1	13	5	18	11	12
Anopheles per 10 bedrooms ...	27	0·3	0	0	0·5	3·3	1·2	9·5	5·7	6·3
Anopheles per 10 bedrooms in adjacent untreated village (Arapa) ...	—	—	42	—	235	—	31	—	65	—

A dramatic fall, almost to zero, is seen in the figures for the daytime catch, which remains low for three or four weeks. After that the catch gradually rises, but is still low 9 weeks after treatment. During that time no infections were found in 50 mosquitos dissected.

Parallel catches in an uncontrolled village, $\frac{1}{3}$ of a mile beyond the spraying zone showed a high output of *gambiae* and *melas* in the area. These findings agree with those quoted above in showing that treatment of houses with DDT may keep the house catch very low for months after treatment.

It remained to be seen whether this low house catch also meant reduced biting activity, and reduced infectivity of mosquitos.

Experiment 2. Behaviour of A. melas *in experimental huts treated with DDT in kerosene.*—These huts have already been described in the introduction, and in the report on pyrethrum experiments. They are constructed in such a way that mosquitos leaving the hut are trapped in a window cage.

Experiment 2a.—One hut was treated with 5 per cent. DDT in kerosene at the rate of roughly 100 mg. DDT per sq. ft., of wall and ceiling surface. In this hut there were no hanging clothes or curtains, and every possible resting place was treated. The numbers of mosquitos in the hut and in the window cage were compared with those of an untreated control. The results are shown in Table V.

TABLE V.

Treatment of experimental huts with DDT. One hut treated at rate of 100 mg. DDT per sq. ft. Other hut untreated. Catches refer to freshly blood-fed *Anopheles* (95 per cent. *melas,* 5 per cent. *gambiae*). Window traps record mosquitos leaving hut after feeding.

		Before treatment	After spraying					
			1 Week	2 Week	3 Week	4 Week	5 Week	6 Week
Number of visits		8	5	5	6	6	3	4
Treated hut	Hut catch	77	0	0	1	0	1	0
	Window trap	27	44	89	176	255	133	178
Untreated hut	Hut catch	91	189	244	390	140	138	194
	Window trap	25	56	12	223	294	123	35

It will be seen that throughout the course of the experiment—6 weeks—the treated hut remained almost completely free of resting *Anopheles*, but the window cage catch showed that as early as the fourth day after treatment, mosquitos were feeding in the hut and leaving after feeding. The number of mosquitos doing this increased steadily, and as early as a week after treatment 20 or 30 *melas* were feeding every night in the hut. All the mosquitos which fed left the hut and were caught in the window trap. Among those mosquitos there was no appreciable mortality in the following 48 hours.

Mats and sheets were spread on the floor of the treated hut, but no evidence was found that any of the mosquitos were succumbing to the insecticide inside the hut.

Experiment 2b.—Another hut was treated with DDT at the rate of 250 mg. DDT per sq. ft. (actually ½ gallon of 2½ per cent. DDT in kerosene for the hut; this is a heavy dose ensuring that every part of the walls and ceiling was soaked with kerosene). The mosquito catches on the following days were compared with the untreated control.

The results are shown in Table VI.

TABLE VI.

As in Table V, but experimental hut treated at rate of 250 mg. DDT per sq. ft.

		Before treatment	After spraying			
			1 Week	2 Week	3 Week	4 Week
Number of visits		5	4	6	5	3
Treated hut	Hut catch	189	0	0	0	0
	Window trap	56	2	28	157	82
Untreated hut	Hut catch	273	196	390	115	138
	Window trap	40	10	223	245	123

The hut remained free of biting *Anopheles* for about 10 days after which increasing numbers started feeding and leaving as before. Again, no dead *Anopheles* were found on the floor matting of the treated hut and the mortality, during 48 hours after feeding, among mosquitos leaving the hut was practically nil.

In both these experiments we could find no evidence of any lethal action of DDT, its only property of note being a marked residual irritant effect, sufficient to prevent mosquitos from resting, but not from feeding, in the treated hut for many weeks after treatment.

It must be borne in mind also that these experiments were carried out under ideal controlled conditions. In the ordinary village there are numerous resting places in houses which cannot adequately be treated with DDT in kerosene, such as bundles of hanging clothes, curtains, wall pictures, and all feeding utensils, the combined effect of which would be to reduce even further the short period of protection.

These experiments still leave unexplained the short period of 4–10 days after treatment, during which no feeding takes place. The following experiments throw some light on this.

Experiment 3. Treatment with kerosene alone.—One hut was treated with kerosene alone in the same quantity as was used in Experiment 2b, *viz.* ½ gallon per hut. The mosquito catches on the following days were compared with those of an untreated hut. The results are shown in Table VII.

TABLE VII.

Experimental huts treated with kerosene alone at rate of ½ gallon to the hut. Effect of treatment on total catch (hut+window trap) of blood-fed *melas*.

	Days before treatment			Days after treatment										
	3	2	1	1	2	3	4	5	6	7	8	9	10	11
Treated hut	43	79	75	0	1	6	0	2	1	—	0	—	4	34
Untreated hut	65	128	59	43	75	51	—	60	—	—	24	—	56	—

It is seen that the heavy dose of kerosene alone produces an immediate drop in the number of feeding mosquitos, and that its repellent effect lasts for over a week.

Another experiment in a different hut, using ¼ gallon kerosene alone, produced much the same effect, both hut and window catches remaining very low for about a week after treatment.

From these experiments it appears that the few days complete protection which follows spraying the hut with DDT in kerosene is mainly if not entirely due to the repellent effect of the heavy dose of kerosene which accompanies the DDT.

Experiment 4. Effect of house spraying with DDT in kerosene on numbers of melas *in outside resting places.*—In the first experiment on the mass spraying of villages, the outside resting places in that particular area were too diffuse to provide any data. In the present experiment we selected an isolated village where we had always been able to make regular collections in particularly suitable outside resting places.

The village houses (70 rooms) were sprayed with 5 per cent. DDT in kerosene at the rate of about 200 mg. DDT per sq. ft. Regular hand catches were made in the houses and in the adjacent outside resting places. The results are shown in Table VIII.

TABLE VIII.

The effect of spraying village houses with DDT on the numbers of blood-fed and gravid *Anopheles* (mainly *melas*) caught indoors and in outside resting places. Makoko village (70 rooms) sprayed with 5 per cent. DDT in kerosene at rate of approximately 200 mg. DDT per sq. ft.

		Week before spraying	After spraying				
			1 Week	2 Week	3 Week	4 Week	5 Week
Rooms searched		15	15	15	10	13	12
Anopheles found in houses		18	0	0	0	0	0
Anopheles found in outside resting places	Total	9	48	75	41	113	53
	Mean per catch	9	16	25	41	37	17

The above figures show again the almost complete elimination of resting *Anopheles* from the treated houses, but the outside catch shows, if anything, an increase as compared with before treatment. Unfortunately, the methods used do not justify a more definite conclusion which the figures seem to suggest. It has not been possible to standardise the outside catch and, although most of the blood-fed and gravid females normally found in outside resting places are those that have left houses after feeding, there is no obvious relationship from day to day between the house catch and the outside catch.

Dissection of 295 of these blood-fed and gravid mosquitos (mainly *melas*), taken in outside resting places beside the treated village, showed 6 gland infections—a sporozoite rate of 2 per cent.—infected mosquitos still being found 5 weeks after the village was sprayed. (The sporozoite rate of 113 *melas* from these outside resting places during the previous year was 2·7 per cent.)

The village is ½ mile from the nearest houses in Yaba and, although the possibility of occasional mosquitos feeding in the town and resting outside in the village cannot be overlooked, our experience has been that most outside resting takes place close to the feeding ground. In view of this it seems fairly certain that the blood-fed and gravid mosquitos caught in the outside resting places were almost entirely composed of those that had fed in the treated houses and left after feeding.

These observations, taken in conjunction with the results of the hut experiments, suggest that the sharp fall in the house catch following treatment with DDT in kerosene does not necessarily mean a corresponding reduction in mosquito population or biting activity. The most obvious effect of the treatment is to bring about a complete shift from indoor to outside resting places. Furthermore, the sporozoite rate of those mosquitos caught outside is such as to suggest that malaria transmission is still possible.

Discussion on Results of House-spraying with DDT.

The experiments described in this paper show that house spraying with DDT in kerosene cannot be regarded as an effective measure against *A. gambiae* and *A. melas* in West Africa. Native houses treated with DDT may remain almost completely free of *resting Anopheles* for many weeks after treatment but, as early as the fourth day after spraying, females can enter the treated houses and leave after feeding. Among the large number of *Anopheles* that leave the house after feeding there is no appreciable mortality in the following 48 hours.

Further proof that *gambiae* and *melas* can feed in a treated hut, and survive unaffected, is afforded by the regular finding of blood-fed and gravid females hiding in outdoor resting places in a village in which all houses have been treated with DDT. The occurrence of gland infections in 2 per cent. of the mosquitos caught outside suggests that malaria transmission is not checked.

At first sight these findings at Lagos appear to contradict completely reports sent in from many other parts of the world. But we consider that the results obtained in the present investigation are unusual only in so far as the methods used were based on a thorough preliminary study of the normal behaviour of *A. gambiae* and *A. melas* in houses. At the moment these findings only refer to these two species in the usual type of native habitation in coastal districts of West Africa. It is possible that these results are due to fundamental differences in behaviour or in reaction to DDT between *gambiae* and *melas* on the one hand, and all other important malaria vectors on the other. But a study of the literature and of the methods used in various countries to study the effect of DDT on adult *Anopheles* in houses shows that in many cases the techniques employed do not preclude the possibility of other species of *Anopheles* escaping unharmed from treated houses.

In the present investigation DDT in kerosene has been shown to have an irritant effect, tending to drive blood-fed mosquitos out of the treated houses, and to make the house no longer attractive as a daytime resting place. The fact that numbers of hungry females may enter and feed in a hut within a week of treatment shows that there is no obvious repellent effect ; furthermore the fact that the number of unfed females caught in the trap is no greater proportionately than in the untreated hut suggests that most hungry females can take their blood meal in the hut without first necessarily coming in contact with the treated walls and roof. As to the exact behaviour of the blood-fed females after feeding, everything indicates that their contact with the walls must be very brief, sufficient to irritate, but not long enough for them to absorb a fatal dose.

With *A. quadrimaculatus*, Say, it has been shown that mortality varies directly with exposure time, but not materially with the dose of DDT applied, provided it is 100 mg. per sq. ft., or more (Fay, Simmons, & Clapp 1945). As to the minimum exposure necessary to bring about a high mortality, opinions tend to differ. For example, Gahan, Travis & Lindquist (1945) found that, with a 15-minute exposure to a surface treated with DDT at the rate of 200 mg. per sq. ft., the mortality of *A. quadrimaculatus* after 24 hours was 46 per cent. Using even shorter exposures, 5 minutes, Metcalf and others (1945) found that with the same mosquito and the same rate of DDT on the surface, the mortality was 20 per cent. on the newly treated surface, and 43 per cent. in the first week after treatment. They found that a 30-minute contact was necessary to give a 100 per cent. mortality up to the second week. With the same species and the same concentration of DDT, Simmons (1945) found a 45-minute exposure to a panel one week after treatment produced 100 per cent. mortality.

34 1?

Kartman & da Silveira (1946) have tried the effect of much shorter exposures on *A. gambiae*, and they found that mosquitos, allowed only 5 seconds contact with a surface treated with DDT at the rate of 125 mg. per sq. ft., showed a 97 per cent. mortality between 10 and 24 hours after contact.

These results show wide discrepancies which cannot entirely be accounted for by specific differences ; but from these results alone it is still possible for a mosquito to be irritated but otherwise unaffected by a very brief contact with a treated surface. Perhaps only a fraction of a second would suffice to prevent further contact and drive the mosquito out of the house. This is possibly what happens with *gambiae* and *melas* in our experiments.

The irritant effect, or activating effect, of DDT in kerosene on mosquitos has also been noticed by other observers such as Metcalf and others (1945), and Knowles &

Smith (1945) on *A. quadrimaculatus*; on *A. albimanus*, Wied., by Trapido (1946), and on *A. darlingi*, Root, by Symes & Hadaway (1945).

The possibility of mosquitos feeding in large numbers in treated houses, and escaping unharmed, has been ignored by many workers, and where the possibility has been realised, all that can be said is that many of the methods used would have given quite misleading results with *A. gambiae* and *A. melas*. For example, in studying the effect of DDT house spraying on various *Anopheles* in the Far Eastern Theatre of War (Medical Advisory Division, 1944) it was considered that by lowering mosquito nets over the catching stations at dawn, mosquitos in treated houses would be trapped before they could leave. With *A. gambiae* and *A. melas* in Lagos, the window cage was found full of active mosquitos, escaping from the treated hut, at least an hour before dawn, and there was every indication that by dawn all the mosquitos which were going to leave the treated house had already done so.

For this reason, also, collecting at 2-hourly intervals through the night—a method used by Symes & Hadaway (1945) with *A. darlingi*—would have yielded misleading results with *gambiae* and *melas* in that most of the mosquitos feeding in the intervals between catches would already have left the house.

Adequate provisions for trapping mosquitos leaving treated houses have been made for *A. quadrimaculatus* by Metcalf and others (1945) by means of exit cones and trap, and the same method has been used to obtain a sample of *A. quadrimaculatus* leaving houses by Simmons and staff (1945). Gahan & Lindquist (1945) consider that most of the *quadrimaculatus* that contact DDT residues in unscreened buildings fly out of open windows and doors, and escape before being knocked down. Metcalf and others (1945) found that among those *quadrimaculatus* trapped leaving treated houses or "nail kegs" there was a mortality of 95–100 per cent. in the following 48 hours. Similar conclusions were reached by Tarzwell & Stierli (1945) and by Gahan & Lindquist (1945).

From observations on *A. gambiae* and *A. melas* in West Africa it appears that the exact nature of the trap is of greatest importance in reaching accurate conclusions about mosquitos leaving huts. Not only has the window trap to act as a direct attraction to mosquitos irritated by contact with DDT in kerosene treated surfaces, but it also had to be an efficient one-way trap, offering no hindrance to mosquitos trying to leave the house, yet at the same time designed so that there is no possibility of mosquitos escaping back into the dark house at dawn. If the mosquitos are artificially prevented from leaving the house by closing doors or windows, it is possible that they may have just sufficient extra contact with the treated walls to absorb a lethal dose of DDT.

While the great majority of African village houses, built of mud walls, and thatch or corrugated iron roof, possess obvious holes and gaps through which "irritated" *Anopheles* can make a swift escape, there are better class houses, particularly in towns, where open exits are not so obvious. This applies particularly to the well-built Creole houses of Freetown and neighbouring villages, and to the African bungalows and villas in Lagos. Such houses usually have glass windows which are kept firmly closed at night, and in these circumstances treatment with DDT might possibly produce a more satisfactory kill, simply because mosquitos flying towards the closed windows in an attempt to leave the house would be trapped in the room long enough to obtain lethal contact with DDT.

It is possible that further research will reveal differences in the effect of house spraying with DDT on different species of *Anopheles*, depending on different habits, different housing and types of wall surface, and also on whether the *Anopheles* are highly efficient vectors like *gambiae, melas, funestus* and *minimus*, or mild vectors like *culicifacies* in South India and *darlingi* in British Guiana (Giglioli, 1946), which maintain malaria transmission by sheer weight of numbers. The present investigation, however, shows that with *melas* at least, and possibly *gambiae*, the treatment of West African village houses with the present form of DDT in kerosene does not produce the dramatic reduction in mosquito population suggested by the house catch. The irritant effect of this particular form of DDT treatment evidently prevents full use being made of the known lethal properties of this insecticide. Until this obstacle has been overcome, the striking reduction in the numbers of mosquitos resting in houses treated with DDT in kerosene cannot be accepted as evidence of effective control.

Summary.

1. The behaviour of *A. gambiae* and *A. melas* in houses is described in relation to possible use of pyrethrum and DDT.

PYRETHRUM.

2. Mass spraying of isolated villages with pyrethrum in kerosene four times a week reduces the day-time catch of *Anopheles* to one-third. The sporozoite rate of mosquitos in sprayed villages remains much the same as before treatment.

3. Observations in experimental huts with window traps attached show that pyrethrum exerts a repellent effect on many hungry *Anopheles* for a day or two after spraying, provided blood meals can be obtained easily in adjacent untreated houses.

4. Where all available huts occupied by man are sprayed, there is no great repellence to hungry mosquitos, which continue to feed in large numbers even in huts sprayed every day.

5. In untreated huts about 20 per cent. of the blood-fed females leave the house at dawn after feeding. In huts sprayed daily the proportion increases to about 80 per cent.

6. The fall in the house catch in villages sprayed 4–6 times a week is not due to any great reduction in the mosquito population, but to a shift from indoor to outside resting places.

7. As intense house spraying with pyrethrum fails to bring about any substantial reduction either in the mosquito population or in their infectivity, it is not likely to be of great anti-malaria value in West Africa.

DDT.

8. Mass spraying of village houses with 5 per cent. DDT in kerosene produces a dramatic fall in the day catch of resting *Anopheles* in houses. The catch remains very low for 4 weeks after treatment and it is still fairly low after 2 months.

9. Observations in experimental huts (fitted with window traps) treated with DDT in kerosene show that mosquitos may enter and feed within a few days of treatment. By the second week after treatment large numbers of *Anopheles* may feed every night in treated huts.

10. There is no evidence of mosquitos succumbing to the effects of DDT in kerosene inside the hut. Nearly all the *Anopheles* feeding in such huts leave after feeding and show no appreciable mortality in the following 48 hours.

11. The DDT in kerosene has a marked residual irritant effect on *Anopheles*, driving them out of the house after they have fed, and preventing mosquitos resting long enough on treated surfaces to absorb a lethal dose of DDT.

12. The few days complete protection from biting mosquitos which follows spraying inside the house with DDT in kerosene, is shown to be due mainly, if not entirely, to the repellent effect of the heavy dose of kerosene which accompanies the DDT.

13. Treatment of all rooms in an isolated village reduced the day catch to nil during 5 weeks after treatment. In outside resting places beside the village, blood-fed and gravid *Anopheles*, of which 2 per cent. had sporozoites in the salivary glands, were taken regularly during this period.

14. The apparent elimination of mosquitos from houses following treatment of rooms with DDT in kerosene is due to a complete shift from inside to outside resting places, on account of the residual irritant, but not lethal, effect of DDT-in-kerosene treated surfaces.

15. The sharp fall in the house catch of *Anopheles* following treatment of West African village houses with DDT in kerosene can not, therefore, be accepted as evidence of mosquito reduction or effective control.

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THE EXTERMINATION OF *GLOSSINA PALPALIS FUSCIPES*, NEWSTEAD, BY HAND CATCHING.

By J. P. Glasgow and B. J. Duffy.
*Department of Tsetse Research, Tanganyika Territory**.

(Plate IX.)

Introduction.

Throughout this paper wherever the term *G. palpalis* is used *Glossina palpalis fuscipes*, Newstead, is meant.

It was known that by hand catching, *G. palpalis* could, under certain conditions be so reduced in numbers as to control human trypanosomiasis (*Trypanosoma gambiense*). This was first shown in Kenya by Symes and Vane (1937) and confirmed in the Sudan by Hunt & Bloss (1945). The conditions under which successful control was attained were streams and rivers carrying low to medium densities of *G. palpalis*. Much of Symes and Vane's work was done on a system of streams carrying a low initial density of *G. palpalis*, and under these conditions they considered they had achieved " eradication ".

Eradication is much more desirable than control. Reconnaissance of Symes and Vane's reclaimed area showed, however, that *G. palpalis* had reappeared in some of the streams from which it had been eradicated (see p. 471). While this was probably because the area was inadequately isolated (by a clearing only 1,050 yards wide) from bush containing *G. palpalis*, it meant that the possibility of eradication was insufficiently proved for a large-scale scheme of reclamation to be based on it. The primary object of the present work was to test again, in an area with more adequate isolation, the proposition that *G. palpalis* can be completely eliminated by hand catching. An incidental object was to remove a focus of sleeping sickness.

Location of the Experiment.

The Sari is a small tributary of the Kuja River, which is in the South Kavirondo district of Kenya and flows into Lake Victoria (see Map). They join about 3 miles above the confluence of the Odielo and the Kuja, the various tributaries of the Odielo being the scene of the work of Symes & Vane (1937). The Sari dries back to a series of pools in the dry months of the year ; on the other hand during rains it is too deep to ford.

It is about 16 miles along the river from the confluence with the Kuja to the main road bridge, and in 1944 *G. palpalis* was found to be present in varying densities over almost the whole of this distance. Above the bridge there was no bush and no tsetse. This infestation was producing a considerable amount of sleeping sickness among the inhabitants of the valley. At the dispensary established about half-way down the valley in September 1944, 230 cases were treated in the remaining months of 1944.

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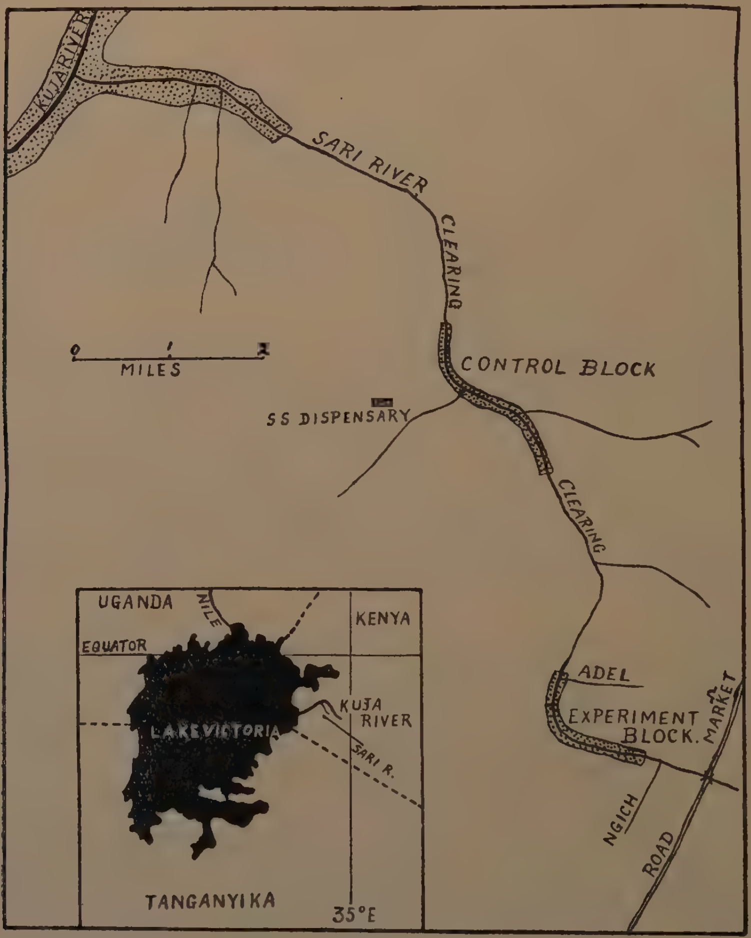
*Pending the re-organisation on an inter-territorial basis of tsetse research in East Africa, it was arranged in 1944 that the Department of Tsetse Research of Tanganyika should extend its operations to Kenya and Uganda and undertake without delay certain field experiments. This paper describes the first of the field experiments to be completed.

S. Napier Bax, Acting Director.

An aerial view of this country would show much open ground with a network of evergreen thicketed streams. The open ground consists of cultivation, grassland and wooded grassland. Vegetational details are given in the appendix. The Sari, with its tributaries, forms an important part of this network (see Plate IX, fig. 1). The thickets on the tributaries of the Sari join, or nearly join, in several instances, the thickets on the tributaries of its parallel neighbours the Oyani, to the west, and the Kuja, to the east. The tributaries of the Sari are normally not infested with *G. palpalis*, but nevertheless light infestations have several times been found on streams previously considered free of *palpalis*, so it would be no surprise to discover an odd one anywhere on this network.


KUJA RIVER
SARI RIVER
CLEARING
0 1 2
MILES
CONTROL BLOCK
SS DISPENSARY
CLEARING
ADEL
EXPERIMENT BLOCK.
MARKET
NGICH
ROAD
UGANDA
NILE
KENYA
EQUATOR
LAKE VICTORIA
KUJA RIVER
SARI R.
TANGANYIKA
35°E

Map 1.

There were only three patches of heavy bush on the Sari: the lower reach which is continuous with the riverine forest of the Kuja; a patch below the Sleeping Sickness Dispensary, and a patch near the main road bridge. Between these patches the bush was light and intermittent and could be completely removed at no great cost.

This light intervening bush was removed and replaced by short grass, thus isolating the Experiment Block, near the bridge, and the Control Block, half-way down the river (see Map 1). These two blocks are separated by a clearing 5,000 yards along the river, which was completed at the end of March 1945. The Control Block is separated from the finger of Kuja riverine forest on the lower Sari by a clearing 6,600 yards along the river, which was first completed at the end of April 1945, and then increased by clearing some of the tributaries entering the Sari in the clearing in October and November 1945. These clearings were, and still are, meticulously maintained. No woody regrowth is tolerated, nor any long reeds, and within 10 yards of the stream the grass is kept shorter than knee high. Cultivation of tall crops such as maize and sorghum has not been allowed.

It was necessary to isolate the Experiment Block to prevent immigrant *G. palpalis* replacing those which were caught. It was considered necessary to isolate the Control Block also, because of the possibility that *G. palpalis* existed on streams such as the Sari only by virtue of constant immigration from the more heavily infested places lower down. If this were so, isolation alone, by stopping immigration, would have caused *G. palpalis* to disappear from both Experiment and Control Blocks. Table I shows that *G. palpalis* has persisted in the Control Block for 16 months since it was isolated at the end of April 1945, and it is believed that it would persist for ever.

TABLE I.

Densities of *G. palpalis* in the Control Block, from fly-rounds.

	Teneral percentage	Non-teneral female percentage	Non-teneral males per 10,000 yards	Total flies per 10,000 yards
1945				
February	22	66	15	56
March	28	59	13	45
April	30	56	10	32
May	19	26	38	64
June	18	38	31	61
July	21	54	20	56
August	18	45	15	34
September	14	42	14	29
October	3	73	4	14
November	4	47	8	15
December	nil	29	10	15
1946				
January	3	53	10	21
February	13	42	7	13
March	34	58	3	11
April	34	40	4	10
May	18	33	10	20
June	17	43	7	16
July	26	36	6	13
August	10	52	7	16

The Experiment Block is 3,800 yards long, measured along the stream. In it, the evergreen riverine bush is quite narrow, perhaps 15 yards on each bank on the average. In places it disappears entirely on one or even both banks, and tongues of riverine bush follow up tributaries for as much as 1,100 yards. There is one small patch of forest in the block, for the rest the bush consists of high thicket with scattered tall trees projecting above it. A detailed description of the vegetation of both blocks will be found on p. 474.

In the Control Block the vegetation consists for the most part of a very thin line of evergreen bush along each bank, but owing to the tortuous course of the stream it has the appearance from a little distance of quite a solid mass of bush. The length of the path which follows the stream is 5,600 yards, but the length of the block in a straight line is much less. In general the bush is highest and widest at the upstream end, lowest and narrowest at the down stream end. There are two small patches of forest and what is probably the remains of a third patch.

There is some evidence, particularly in the Experiment Block, that the bush is in an unstable condition. For instance there are no mature specimens of *Rauwolfia natalensis*, Sond., although mature specimens of this tree are found in the Control Block, and the majority of the *Albizzia* in the Block are obviously immature. Only one full-grown *Kigelia aethiopica*, Decne, was seen, with at least two young ones. This instability is possibly the result of less intensive human use of the valley following the appearance of sleeping sickness.

The Experiment.

(a) *Initial Densities of* G. palpalis.

Fly-rounds, carried out weekly by parties of 3 fly-boys provided with hessian screens, were started in both blocks in February 1945, and showed that the two blocks had in the beginning much the same densities. Tables I and II give the details of the catches. The teneral flies referred to in the tables are those which have not yet fed. The columns in the tables headed "Non-Teneral Female percentage" are of interest because, although it is normal to find 50 per cent. of teneral flies are female, often only 10 per cent. or less of non-teneral flies in catches of undisturbed populations are female. This applies to several other species of *Glossina*, and to catches of *G. palpalis* made on the shore of Lake Victoria. The reason for the high non-teneral female percentage on our streams (it is an observation not confined to the Sari) is obscure, but it is obviously easier to exterminate a population in which females are caught readily than it is to exterminate one in which they are caught with difficulty.

TABLE II.

Densities of *G. palpalis* in the Experiment Block, from fly-rounds.

	Teneral percentage	Non-teneral female percentage	Non-teneral males per 10,000 yards	Total flies per 10,000 yards
1945				
February	34	68	13	62
March	23	67	15	58
April	35	63	17	71
May	78	100	nil	6
June	80	100	nil	3
July	67	0	0·6	2

Note : Hand catching began on 1st May. In August and September fly-rounds failed to produce any tsetse, and were thereafter discontinued.

(b) *Hand-catching.*

Organised hand catching began in the Experiment Block on 1st May, 1945. In all 30 fly-boys were used, organised in teams of 3. In each team 2 men carried between them a hessian screen, while the third carried a small wire gauze box, in which the flies were put, alive, as they were caught. The value of these practices was uncertain, but they were adopted as there was some evidence that they were advantageous. Recently, in a 48-days trial during which screens were used every other day, no increased catch from carrying a hessian screen was recorded. A similar trial of the boxes is not yet complete.

Eight of the 10 teams were used in the main part of the Experiment Block. Four teams made up the morning shift, which worked from 7 till 3, and 4 teams made up the afternoon shift, which worked from 11 o'clock till dusk, so that for about 4 hours each day there were 8 teams working in the block. There were paths cut on both banks of the river, as near the water as possible, and at all times the same number of parties on each bank. Schedules were set out, and fairly well adhered to, which kept the teams spread out over the block as evenly as possible.

The other two teams worked one in each of the two tributaries, the Adel and the Ngich, morning shift only. A second, afternoon, shift was started in the Ngich in September.

The catchers did not work on Sundays. On this day the fly-round was carried out. It was hoped to use the fly-round data as a check on the progress of the work, but as Table II shows, the fly-round figures fell to zero almost as soon as catching-out began.

TABLE III.

Extermination of *G. palpalis* in the Experiment Block.
Total catches of morning and afternoon shifts, with catches on Adel and Ngich included.

	Totals			Teneral			Non-teneral		
	Males	Females	Both	Males	Females	Both	Males	Females	Both
1945									
May ...	122	160	282	56	62	118	66	98	164
June ...	47	37	84	33	24	57	12	15	27
July ...	19	14	33	13	8	21	6	6	12
August ...	0	2	2	0	2	2	0	0	0
September	3	1	4	0	0	0	3	1	4
October ...	1	0	1	0	0	0	1	0	1
Totals ...	192	214	406	102	96	198	88	120	208

Table III gives the actual numbers of *G. palpalis* taken. Of the total, 358 were taken in the main block, the remainder in the tributaries. Of the 358, 233, or 65 per cent., were caught by the morning shift. The block is divided into 4 sections, each 1,000 yards, except section D which is at the downstream end of the block and is only 800 yards long.

TABLE IV.

Distribution of catches in different sections of the Experiment Block.

	A	B	C	D	Ngich	Adel
1945						
May	77	21	134	27	10	13
June	17	1	43	11	7	5
July	6	2	10	5	4	6
August	1	1	0	0	0	0
September	1	0	0	0	3	0
October	0	0	0	1	0	0
Totals	102	25	187	44	24	24

Table IV gives the distribution of the gross catches of *G. palpalis* in the different sections. The Ngich is a tributary some 1,500 yards long which joins the Sari above the Experiment Block, but so close to it that it had to be treated as part of the block.

(c) *Efficacy of the Clearings.*

Many reconnaissances were made in both clearings, but no *G. palpalis* were ever caught in them, except a few which penetrated a couple of hundred yards into the bottom end of the clearing which separates the Control Block from the Kuja riverine forest.

Large numbers of flies have been marked in the Control Block to see whether interchange of flies between this block and other places occurs. In the 15 months beginning with May 1945 the following numbers were marked: 152, 92, 181, 367, 425, 273, 84, 70, 78, 32, 41, 28, 40, 29, 16. After 13.vii.46 no more flies were marked. During the course of hand catching no marked flies were recovered in the Experiment Block (although one was seen in April 1946, see below), and this, in conjunction with the absence of flies in the upper clearing was regarded as being as good evidence as it was possible to get of the effectiveness of the isolation of the Experiment Block.

In August 1945 some *G. palpalis* marked in the Control Block were recaptured in the Kuja riverine forest. At the same time *G. palpalis* was discovered in small numbers in several small tributaries of the Sari entering the clearing between the Control Block and the Kuja riverine forest. Flies had previously been looked for and not found in these tributaries. It was concluded that the thickets on these tributaries were serving as stepping stones for the *G. palpalis*, and the thickets were all removed, since when (September 1945) no more marked flies have been recovered outside the Control Block. At the same time, similar tributaries in the clearing between the Experiment and Control Blocks were cleared as a precaution.

In spite of the encouraging results recorded above, it was recognised that odd *G. palpalis* might arrive in the Experiment Block, not necessarily from the Control Block, but perhaps via the thicket network described above (p. 467). In fact, in April 1946 a fly was seen in the Experiment Block which had been marked in the Control Block 2 or 3 weeks before. The hope that isolation was effective rested in the belief that such odd flies arriving singly could not repopulate the block. This belief is vindicated (see below).

To forestall a possible misconception, it must be recorded here that it is not considered that a 5,000 yard clearing will always stop *G. palpalis*. It is merely reported that this particular clearing, which is very carefully maintained, has stopped *G. palpalis* so far. It is believed possible that even with careful maintenance of the clearing the Experiment Block would sooner or later be repopulated. Now that the Control Block has fulfilled its purpose (demonstrating that the disappearance of *G. palpalis* from the Experiment Block was not spontaneous) it too will be freed of *G. palpalis* by hand catching. Afterwards maintenance of the upper clearing can be relaxed, but not maintenance of the lower clearing, until *G. palpalis* has been removed from the Kuja.

(d) *No Reinfestation.*

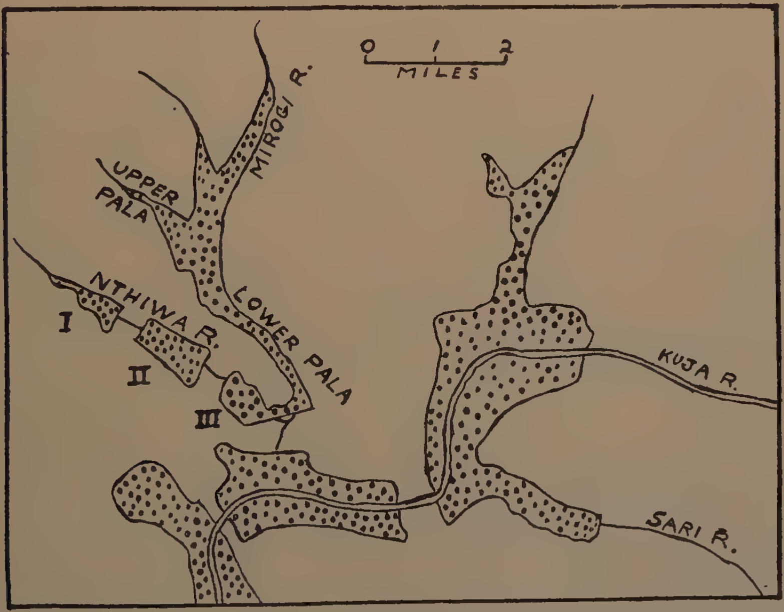
At the end of December 1945 two months had passed without any *G. palpalis* being caught. Hand catching was, therefore, regarded as complete, and the catchers were removed. To give early warning of any reinfestation, however, a party of 3 was kept working in the block, 4 days a week in the main block and 1 day a week in each of the tributaries. This party caught 1 non-teneral male *G. palpalis* in January 1946.

On more mature consideration it was concluded that this party was a mistake. Reinfestation might come from multiplication of a minute residual population which had somehow escaped the catchers, or from the odd immigrant, the possibility of whose arrival had to be admitted. In either case, the presence of 3 catchers might be sufficient to prevent reinfestation. If they caught 1 or 2 flies a month (as they did catch 1 in January) nobody could tell whether those 1 or 2 flies, if not caught, would have died without issue or multiplied exceedingly. Accordingly it was decided to leave the block entirely alone, to give the *G. palpalis* every chance, until June. To give warning of any gross reinfestation a visual reconnaissance (*i.e.* without nets) was done once a month. During each of the March and April reconnaissances one *G. palpalis* was seen, and the April fly had been marked, as already mentioned.

On 20th June, 1946, the hand catching organisation was re-introduced. Only the main block, and only morning shift, was worked, so that only 14 fly-boys were used, as against 50 in the original catching-out. These 14 boys patrolled the block until 5.vii.46. During these 14 working days no tsetse were caught. It is inconceivable that 14 men in 14 days should fail to find any *G. palpalis* if any were present.

(e) *Observations on the Area reclaimed by Symes & Vane.*

The reclaimed area of Symes & Vane (1937) consists of 3 streams, the Nthiwa, the Mirogi and Pala (Map 2). The Mirogi runs into the Pala, which is called Lower Pala below the junction, Upper Pala above it. Table V gives the results of fly-rounds carried out on these streams once a week for the first 8 months of 1946. The Nthiwa fly-round taps only the lower part of the stream (Blocks II and III) because it had been found in 1945 that the upper part (Block I) was free of *G. palpalis*.



Map 2.

The fly-rounds show that reinfestation has not been very serious. Only on the Mirogi does there seem to be an infestation which may be serious, and even this is certainly far below what it was before the operations of Symes and Vane. The Upper Pala and Blocks I and II on the Nthiwa are completely dry for long periods every year, whereas the other streams contain permanent pools. Permanent water therefore seems to be necessary for reinfestation, although not for the original infestation, for Symes and Vane caught many flies in both of these streams.

The observation that reinfestation has been so incomplete on these streams, which appear very open to reinfestation, encourages the belief that hand-catching is a very useful method of reclamation.

TABLE V.

Fly-rounds in the reclaimed area of Symes & Vane.
Densities of *G. palpalis* in terms of non-teneral males per 10,000 yards.

	Nthiwa		Mirogi	Upper Pala	Lower Pala
	Block II	Block III			
1946					
January	0	0	4·4	0	2·6
February	0	0	1·8	0	2·0
March	0	3·8	3·7	0	4·6
April	0	4·7	8·9	0	2·6
May	0	0	6·0	0	2·8
June	0	2·3	5·5	0	5·2
July	0	2·3	4·4	0	2·6
August	0	1·9	5·5	0	0·6

General Considerations.

The methods described above differ from those employed in the pioneer work of Symes & Vane (1937) in two important details: firstly the much greater emphasis placed on preliminary isolation of the area treated, secondly the much larger number of fly-boys used. These changes were introduced with the idea of achieving complete elimination of *G. palpalis* as rapidly as possible.

In round figures, the total catch in a block 4,000 yards long was 400 *G. palpalis*. The April 1945 density of 17 non-teneral males, or 71 " total flies ", per 10,000 yards, may therefore be equated with a total catch of 1,000 flies per 10,000 yards, when hand catching is carried out at such a speed that it is virtually complete in 3 months. The actual flying population on the first day of catching would be perhaps half this, since half the total catch was teneral, and most of these teneral flies were derived from pupae deposited before hand catching had commenced.

It is a common observation (*cf.* for example Gibbins (1941)) that *G. palpalis* is seldom found far from water. This might lead one to suppose that the equation of an apparent density of 17 non-teneral males with a total catch of 1,000 flies would always be true irrespective of the width of the riverine bush, but it is doubted if this is the case. Symes & Vane (1937) removed from their Block II, which is 2,096 yards long and about 200 yards wide, 5,079 *G. palpalis* in 3 months. This corresponds to 25,000 flies per 10,000 yards or an initial density, by the above equation, of 425 non-teneral males per 10,000 yards. It is not known what the initial density in Block II was, but it was certainly less than 425, for this is a density which is seldom reached even on the Kuja, which carries higher densities than its tributaries, both in the writers' experience and in that of Symes & Vane.

It appears then that wide blocks (width measured at right angles to the course of the stream) support a higher population of *G. palpalis* than do narrow blocks, but that this higher population is not necessarily reflected in a higher apparent density.

Conclusions.

(a) *Practical Application.*

It has been demonstrated above that, on streams with narrow bush and a light infestation of *G. palpalis*, hand catchers at the rate of 6 per 1,000 yards of stream eradicate the *G. palpalis* in 3 or 4 months. By "light infestation" is here meant an Apparent Density of less than 50 non-teneral males per 10,000 yards.

There is no reason to doubt that the measure would be equally effective on streams with wide bush and a light infestation of *G. palpalis*, although more catchers, or more time, might be needed.

This is much cheaper than clearing. Moreover, as the first effect of the catchers' activities must be to shorten the average life-span of the flies, it is probable that the first month sees the removal of all flies old enough to have infective trypanosomes. This, however, has not been demonstrated in the field.

These two factors, cheapness and speed, particularly speed in attaining the first result desired (freedom from new cases of sleeping sickness), are the two great advantages of hand catching over any other known method of reclaiming such streams from *G. palpalis*.

The disadvantages are :—

(1) The fly-catchers require more supervision than bush-felling labourers, so that a Field Assistant who could look after 500 bush-clearers might be able to manage only 150 fly-catchers.

(2) The possibility of reinfestation. This is obviously considerable in our experimental area, but in a larger scheme embracing a whole river system it would be much less. Alternatively in some places the cost of hand catching might seem so low that it could be repeated every year or so.

A scheme for eliminating *G. palpalis* from the neighbouring Oyani River, based on the experience on the Sari, has been accepted by the Government of Kenya. A long clearing is to be made at the bottom end where the Oyani enters the *palpalis*-infested Kuja. Elsewhere, where the bush is thin it is to be removed, and where it is heavy *G. palpalis* is to be caught out. The criterion for deciding whether a particular stretch is to be cut out or caught out is cost, and usually there is no difficulty in reaching a decision, as the riverine vegetation falls naturally into two classes—thicket which would be expensive to clear but relatively cheap to catch out, and tenuous bush which can be entirely removed at no great cost. This tenuous bush, although possibly not capable by itself of supporting *G. palpalis* all the year round, harbours sufficient flies, which are derived from the patches of heavier bush, to transmit sleeping sickness.

(b) *The Value of the Bush.*

One of the advantages claimed for hand catching as a method of *G. palpalis* reclamation has been that it saves valuable bush, thereby preventing desiccation and erosion. However, the bush that has been saved on the Sari is of little value either to the forester or to the African peasant, who is in this area increasingly relying on his plantations of quick-growing straight *Eucalyptus* for building materials and fuel. Its rôle in preventing desiccation and erosion is debatable. Staples (1941) found that under some conditions streamside trees use more water by transpiration than they save from evaporation by their shade. It is obvious enough on the Sari as elsewhere that bush does not prevent the erosion associated with places where cattle water regularly. The alluvial flats on which the bush grows are the best crop land in the valley, and for this reason the bush is in any case doomed (unless actively protected) in the face of the expanding population which may be expected to follow sleeping sickness reclamation.

For these reasons the advantages of hand catching as a method of *G. palpalis* reclamation have been assessed in terms of rapidity and cheapness, and have laid no stress on saving the bush. A new freedom is, however, conferred on those who plan better land utilisation. Such riverine bush can be saved, modified or removed without reference to *G. palpalis*.

Vegetational Details.

Away from the Streams.

The country is open and rolling, the ridges being 500 to 1,000 feet above the valley bottoms. The altitude of the bottoms is about 4,000 feet above sea level, and of the highest ridges about 5,000 feet. There is much cultivation, the chief crops being maize, sorghum, *Eleusine*, sweet potatoes, cassava and sesame. The people keep cattle, sheep and goats. Much of the grassland is lightly wooded, the wooding consisting chiefly of at least two shrubby species of *Combretum*, and on the ridges a tree, *Combretum gueinzii*, Sond. subsp. *splendens* (Engl.), Exell. Other important trees are a *Ficus* sp., *Bauhinia thonningii*, Schum., *Acacia macrothyrsa*, Harms. The people plant hedges of *Euphorbia tirucalli*, L., round their homesteads, and many have small plantations of *Eucalyptus* sp. or *Markhamia platycalyx* (Bak.) Sprague. The rainfall is about 50 inches a year, tending to come in two wet seasons in May and November, but it is usual to have some rain every month. The first three months of the year are the driest and hottest.

Experiment Block.

The block is arbitrarily divided into 4 sections, A, B, C, D, containing respectively 1,000, 1,000, 1,000, 800, yards of river. Section A is upstream and D downstream.

Entering *Section A* at the top (Plate IX, fig. 2) there is a stretch of dense regeneration thicket containing :—

Allophylus africanus, P. Beauv., *Cassia didymobotrya*, Fresen, *Clausena anisata* (Willd.), Oliv., *Dombeya dawei*, Sprague, *Grewia* sp., *Oncoba spinosa*, Forsk., *Phoenix reclinata*, Jacq., and *Triumfetta tomentosa*, Boj. and coppicing stumps of the tree *Sapium ellipticum* (Hochst.) Pax.

Following this there is a stretch of about 100 yards of closed canopy forest. The trees of this forest are mostly *Sapium ellipticum* with a few *Erythrina sereti*, De Wild. One of the *Erythrina* has been killed by an epiphytic fig which is now very large. The thicket beneath these large trees is composed of much the same constituents as those noted above. This is the only part of the block where the high canopy is closed—elsewhere, though large trees may be many, their crowns do not touch (see Plate IX, fig. 3).

The remainder of *Section A*, after the two *Erythrina sereti* shown in the photograph, consists of scattered *Sapium ellipticum* with a few *Erythrina sereti* and one *Kigelia aethiopica*, Decne (the only mature specimen of this tree in the block). The undergrowth is as before with the addition of some immature *Rauwolfia natalensis*, Sond.

In *Section B* the large trees tend to be smaller. *Sapium ellipticum* is the commonest, with some *Albizzia* sp. Only one *Erythrina sereti* was noted.

In *Section C* there are a number of large epiphytic figs, with *Sapium ellipticum* and *Albizzia* sp. (see Plate IX, fig. 4). There is one *Pachystela brevipes* (Bak.) Engl., a tree which is common on the Kuja but rare here.

In *Section D* the commonest large tree, as elsewhere, is *Sapium ellipticum*. There are some fairly large *Ficus vallis-choudae*, Del., and some small epiphytic figs. There are a number of immature *Albizzia coriaria*, Welw. ex Oliv. and immature *Albizzia zygia*, Macbride. At the edge of the bush (*i.e.* away from the river) are some *Entada abyssinica*, Steud. On the Adel tributary which enters this section there are some immature *Rauwolfia natalensis*, Sond. The course of this stream is conspicuous from a long way off as a fine row of *Sapium ellipticum* (see Plate IX, fig. 5.)

In general, other constituents of the undergrowth more or less generally distributed are *Maesa lanceolata*, Forsk., *Ochna macrocarpa*, Engl., *Trimeria tropica*, Burkill, and *Vernonia* sp.

A small tree, widely distributed but not very common, is *Bridelia micrantha* (Hochst.) Baill. Its timber has a high reputation for its resistance to termites.

In the stream a low sedge (Cyperaceae) is common, as is *Sesbania aegyptiaca*, Poir., *Mimosa asperata*, L., and *Polygonum lanigerum*, R. Br.

Throughout the block *Sapium ellipticum* is the commonest large tree, and *Allophylus africanus* is the commonest member of the undergrowth.

Control Block.

The Control Block is divided into 4 sections, A, B, C, D, of roughly equal lengths, Section A being downstream and D upstream.

At the bottom end of *Section A* there is a stretch with a single line of trees on each bank. The trees are small coppiced specimens of *Rauwolfia natalensis*, *Allophylus africanus*, *Grewia* sp., *Bridelia micrantha* (Hochst.) Baill., and *Markhamia platycalyx* (Bak.) Sprague.

In the next stretch one or both banks are covered with dense thicket composed of all the above species together with *Oncoba spinosa*, Forsk., and others. Occasional specimens of *Rauwolfia natalensis* and *Albizzia zygia*, Macbride, stick out above the top of the thicket.

Section B begins with a narrow thicket, a continuation of that just described, with the addition of *Dombeya dawei*, *Triumfetta tomentosa* and *Vernonia amygdalina*, Del. Then a small tributary enters the left bank. This tributary carries many fine *Acacia campylacantha*, Hochst., some of which stand on the bank of the Sari. From here on the vegetation is denser and there are more large trees—*Ficus vallis-choudae*, Del., larger *Albizzia zygia* and large *Rauwolfia natalensis*.

Section C begins with a stretch of low dense thicket, followed by a patch of large trees—*Sapium ellipticum*, *Albizzia zygia*, *Bridelia micrantha*. After this comes a stretch of higher thicket with occasional *Acacia campylacantha*. Then the path passes through a tunnel of *Allophylus africanus*, *Grewia* sp. and others to another patch of forest composed of *Sapium ellipticum*, *Rauwolfia natalensis*, *Ficus* sp. and *Albizzia zygia*. A stretch of high thicket is followed by a small patch of *Ficus vallis-choudae* with *Rauwolfia natalensis*. After a natural open space there follows a narrow strip of tallish bush composed of *Ficus vallis-choudae* and another, epiphytic, *Ficus*, *Markhamia platycalyx*, *Rauwolfia natalensis*, *Bridelia micrantha*, *Grewia similis*, K. Sch., *Allophylus africanus*, *Sapium ellipticum*, *Albizzia zygia*, *Bersama holstii*, Guerke, and *Euclea lanceolata*, E. Mey.

In *Section D* the same narrow bush continues with the addition of *Acacia* sp. (? *kirkii*, Oliv.) and *Oncoba spinosa* to a banana garden round which a number of *Markhamia platycalyx* and two *Grevillea robusta* (an exotic) are growing. By this garden is an enormous spreading *Sapium ellipticum*, over 100 feet high. From here the same narrow tallish bush continues to the end, and also lines an old disused bed of the Sari which still holds water. What with this old bed and the convolutions of the Sari itself, the end of the block, as seen from the clearing above it, appears about 300 yards wide.

The tree most characteristic of the Control Block is *Rauwolfia natalensis*, a tree with a wide dense crown and a superficial resemblance to a mango.

Summary.

Two blocks were isolated by 3- or 4-mile clearings on a small river with a narrow strip of riverine bush and a low density of *Glossina palpalis fuscipes*.

Catchers at the rate of 6 per 1,000 yards of stream caught almost all the *G. palpalis* in one block in 3 months and all in 6 months.

Eight months later there was no reinfestation.

All this time *G. palpalis* persisted in the Control Block.

An area from which *G. palpalis* was removed by hand catching 10 years ago was examined. It is concluded that it is possible not only to hand catch all the *G. palpalis* but also to avoid reinfestation.

Hand catching is the quickest and cheapest known method of reclaiming streams of this type from *G. palpalis*.

Vegetational descriptions are given.

Acknowledgements.

We have already recorded that this work is in effect a continuation of that of Symes & Vane (1937). If in places we appear to criticise their work, we are aware that they were always short of money, whereas, thanks to the Colonial Development and Welfare Fund grants, we were not hampered by this factor in designing our experiment.

Mr. Napier Bax, Acting Director of Tsetse Research, Messrs. Potts and Whiteside, and Dr. Jackson, of the Department, and Dr. Garnham, of the Kenya Division of Insect-Borne Diseases, have helped us a great deal with discussions and correspondence. Mr. P. J. Greenway, Systematic Botanist at the East African Agricultural Research Institute, has identified our botanical collections ; without this help the botanical appendix could not have been compiled. Finally, we must record our indebtedness to the thorough and careful work in the field of our African assistant Mr. Haggai Onono.

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APPENDIX.

During 1946, 65 persons contracted sleeping sickness in Sakwa location, through which the Sari river flows, and the evidence suggested that they were infected in the Control Block. The work of removing *G. palpalis* from this block was therefore begun without delay, in December 1946. The work was done in exactly the same way as has already been described for the Experiment Block, except that the catchers were now used in teams of two without a screen, instead of teams of three with a screen as formerly. Table VI summarises the progress of the extermination, and Table VII gives the distribution of the gross catches of *G. palpalis* in the different sections of the block. Regular searches were made also in four thicketed tributaries which run into the Control Block, but no tsetse were found.

TABLE VI.

Extermination of *G. palpalis* in the Control Block.
Total catches of morning and afternoon shifts.

	Totals			Teneral			Non-teneral		
	Males	Females	Both	Males	Females	Both	Males	Females	Both
1946									
Dec. ...	28	39	67	12	9	21	16	30	46
1947									
Jan. ...	13	28	41	8	19	27	5	9	14
Feb. ...	2	3	5	1	0	1	1	3	4
March ...	1	2	3	1	1	2	0	1	1
April ...	0	2	2	0	2	2	0	0	0
Totals ...	44	74	118	22	31	53	22	43	65

TABLE VII.

Distribution of catches in different sections of the Control Block.

	A	B	C	D
December 1946 ...	8	45	9	5
January 1947 ...	6	30	2	3
February	1	4	0	0
	15	79	11	8

In May 1947 the catching-out organisation was re-introduced into the Experiment Block. The block was patrolled for 6 days (12th–17th May) by eighteen trained catchers (transferred from the work in the Control Block). No tsetse were caught or seen. This was 11 months after the last examination of the block (p. 470) and 17 months after extermination was completed in December 1945.

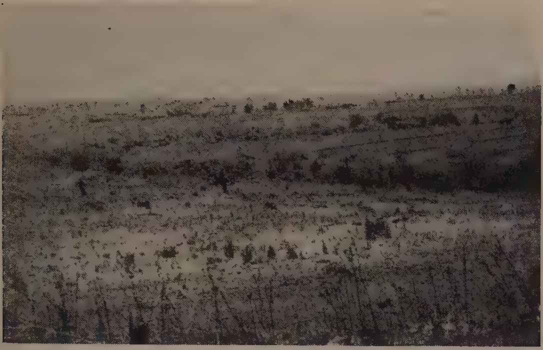

Fig. 1.

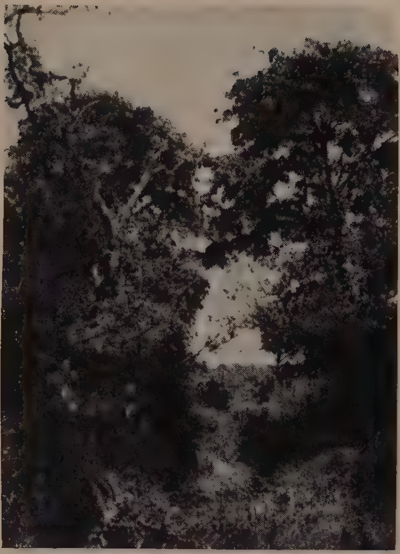

Fig. 3.

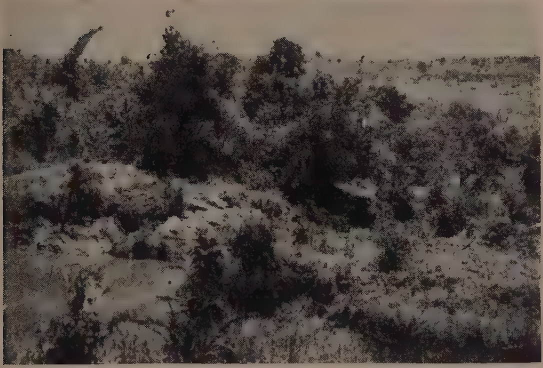

Fig. 2.

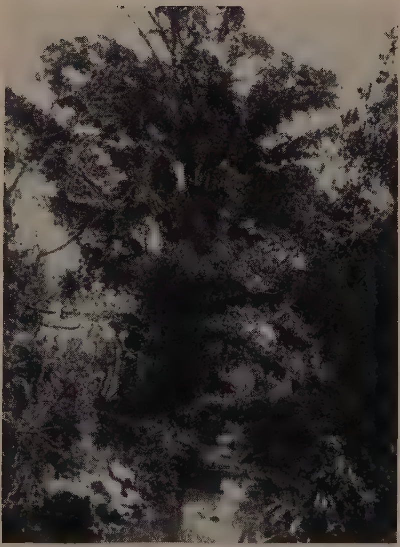

Fig. 4.

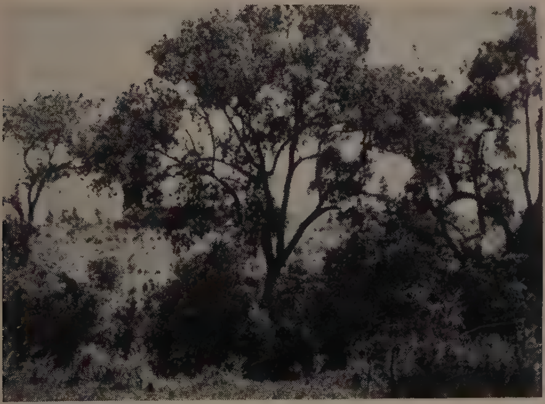

Fig. 5.

Fig. 1. View of the Sari valley looking towards Wendo market. The thicketed stream nearest to the camera is an uninfested tributary.

Fig. 2. Looking at the top end (Section A) of the Experiment Block, from the clearing above it.

Fig. 3. Two *Erythrina sereti* De Wild., outliers of the patch of forest in Section A of the Experiment Block. They stand one on each bank of the Sari.

Fig. 4. One of the smaller of the epiphytic figs in Section C of the Experiment Block. This one is about 40 feet high.

Fig. 5. The Adel stream, *Sapium ellipticum* (Hochst.) Pax, 60 feet high, overhanging the narrow thicket.

SANDFLY CONTROL WITH DDT RESIDUAL SPRAY. FIELD EXPERIMENTS IN PALESTINE.

By F. Jacusiel, M.D., D.T.M. & H.
Late Major R.A.M.C.

L.C.

Very little work has been carried out on the control of sandflies with DDT and still less has been published. The neglect of the use of DDT in this field seems unfortunate, as sandfly-borne diseases play an important rôle in the life of the civil population and military personnel in large areas of the tropics and sub-tropics. In the civilian population visceral and cutaneous leishmaniasis are the main problems involved. In armies, sandfly fever with its rapid and widespread outbreaks in non-immune troops can seriously hinder or even totally impede military operations—several such instances occurred during the past war in the Mediterranean theatre—and an efficient method of controlling sandflies would prove a most valuable addition to military preventive medicine.

Furthermore, although previous attempts to control these insects have proved unsuccessful, the few tests so far carried out with DDT residual spray have given more than satisfactory results. The control of sandflies and sandfly-borne diseases has, in the past, been unsuccessful for two main reasons :

(1) No practicable method of destroying the larval stages has yet been found, and the abolition of potential breeding grounds is, in the majority of cases, far too costly and often impossible.

(2) Protection against the adults requires screens of bed-nets of very narrow mesh, a great disadvantage in hot climates, which constitutes a serious obstacle to their conscientious use.

The only account of sandfly control work with DDT so far published is that by an American Army team (Hertig & Fisher 1945) who conducted a series of field tests in Palestine in 1944. These scientists worked in close contact with the author and their observations served as a basis for the tests described below.

The great effectiveness of DDT residual spray against sandflies was clearly demonstrated in general in the 1944 field tests but a number of important points were left open and needed further investigations :

(*a*) The period of observation by Hertig on the residual effect was limited to 24 days, after which the sandfly season had come to an end. The DDT film was then still fully efficient, so that the maximum length of residual effect remained to be determined.

(*b*) The only species of *Phlebotomus* present in the 1944 tests (Jordan Valley near the Dead Sea) was *P. papatasii*, Scop. It seemed, therefore, desirable to carry out further tests in a locality harbouring also other species, especially of the *major* group, in view of their importance in the transmission of visceral leishmaniasis.

(*c*) Breeding conditions in the semi-desert of the Dead Sea area are peculiar and rather different from those commonly encountered. For the common situation of breeding places closely associated with houses (rubble, old stone-walls, cracks in masonry, etc.) specially adapted methods of application of DDT residual spray appeared to be indicated, methods which could not be tried out under the local conditions of the 1944 tests.

The field tests described below were designed to elucidate these and other points during the 1945 sandfly season.

Experimental Work.

The experiments were conducted, from August to October 1945, in the village of Rosh Pina, in the Galilee district of Northern Palestine, one of the oldest Jewish colonies founded more than 60 years ago. It consists of some 40–50 houses drawn out along the slope of a hill, the highest part being 480 m., the lowest 380 m., above sea level.

Most houses are old (40–60 years), surrounded by courtyards and various outbuildings (stables, fodder and grain stores, etc.) and boundary walls of rough stone. Roughly hewn stone is the main building material. Sandfly breeding places abound in the immediate vicinity of the houses, consisting chiefly of damp corners in the courtyards and crevices in the old and weathered stone walls. Rooms in the houses have rough whitewashed walls, the ceilings often being constructed of wooden beams into the crevices of which sandflies creep during daytime. Average dimensions of rooms are 3·60 m. × 4·80 m. × 3·20 m.

During the period of the tests, maximum temperatures ranged from 24·6 to 37·5 C., with a mean of 31·5 C., minimum temperatures from 14·4 to 26·6 C., with a mean of 20·2 C., and relative humidities varied from 12 to 70 per cent.

These data are recorded because the length of residual effect of DDT films depends to some extent on meteorological conditions ; thus the higher the temperature the shorter the residual effect.

Sandfly Population.

Some catches and night observations were made during July and regular counts were started on 6th August, 9 days prior to the first application of DDT.

Catches and counts were made in different parts of the village at night from 8 p.m. onwards (sometimes until after midnight) and in the morning from 6 to 8.30 a.m., both in rooms and outside on walls.

Specimens were either dissected fresh in saline or preserved in alcohol and later cleared and identified.

The three prevalent species were *P. papatasii*, Scop., *P. major*, Annan., and *P. chinensis* var. *simici*, Nitzulescu. A single male specimen related to *P. sergenti*, Parrot, has been described as a new species (*P. jacusieli*) by Theodor (1947).

All three common species were found in every part of the village, but the proportions varied, i.e. certain species were abundant in some and rare in other parts of the village. Thus, evening catches on outside walls in a courtyard in the upper village always gave mainly *P. major* with very few *P. papatasii*, while catches in and around one particular house in the lower village always produced a high proportion of *P. chinensis*. The habits of *P. papatasii* and *P. major* differ somewhat as *P. major* is found more frequently outside and for longer periods of the night than *P. papatasii*, and the proportion of *P. major* inside houses is higher in the evening than in the morning. This is due to the pronounced negative phototaxis of *P. major*, which in the morning either creeps deep into cracks and crevices or leaves the house altogether for dark hiding places outside. This is illustrated by Table I.

TABLE I.

Distribution of sandflies in different places and at different times.

	In houses (evening)	Percentage	In houses (morning)	Percentage	Outside (evening)	Percentage
P. major...	15	35·7	7	29·2	9	47·4
P. papatasii	21	50	16	66·6	3	15·8
P. chinensis	6	14·3	1	4·2	7	36·8
Total...	42	100	24	100	19	100

The figures given in the table are too low to draw definite conclusions on the bionomics of the various species but they confirm similar observations by other authors (Adler & Theodor, 1931).

Some observations on the peculiar habits of *P. chinensis* are described on p. 485.

The following figures give a general idea of the sandfly density in Rosh Pina :—

Average count of sandflies in a single room	17
Maximum count in a single room	126

Figures refer to 25 rooms examined prior to DDT spraying.

Technique.

After preliminary daily counts in various parts of the village (6th–13th August), rooms and houses were selected for the experiments. The rooms showing the highest counts were chosen for treatment with DDT, in order to make the test more stringent. Other rooms were used for comparison as untreated " controls ".

In each part of the village, with its particular distribution of species, five test groups were arranged consisting of a number of treated and untreated rooms in one or more houses, and are referred to below as group A, B, C, D & E, respectively.

Spraying was carried out with a hand-operated sprayer of the " direct pressure " type (Smith-Banner model). The standard dose used was 1 gal. (4·5 litre) of 5 per cent. DDT in kerosene per 100 m², i.e. 2 grammes (g.) DDT per m² (200 mg./sq. ft.). In some rooms half the amount (1 g. per m²) was given by using the same amount of a 2·5 per cent. DDT solution. The percentages of DDT in solution are given in terms of p′ p′ DDT.

Two different spraying procedures in rooms were tried. In one, walls and the whole ceiling, in the other, walls and a foot-wide strip of the ceiling along the wall-ceiling junction only, were sprayed. In group C, outside walls were treated with 2 g. per m².

The results of the tests are summarised in Table II.

Figures represent the mean of two counts, evening plus morning count divided by two, within 24 hours.

In some instances the figures represent a single count in 24 hours, where one of them could not be made for technical reasons. Nil counts are marked by the word *Nil*, while blanks on some dates denote " no observation made in the particular group ". These gaps are an indication of the technical difficulties encountered, such as locking of rooms or houses during temporary absence of inhabitants.

In each group the figures are the sums of counts in *all* treated rooms and *all* control rooms respectively.

TABLE II.

Effect of spraying with DDT on sandfly counts in rooms.

Test Group ...	A		B		C		D		E		F
Date	Con-trols	Test rooms	Con-trols	Test rooms	Con-trols	Test rooms	Con-trols	Test rooms	Con-trols	Test rooms	Con-trols only
6th August	32	90			65	17	16		10		
7th ,,	65	119			33	28	16		12		
8th ,,	50	116			34	52	20	29	10	4	
10th ,,	42	112			52	56	46	46	13	4	
11th ,,	26	83			1	15	36	29	5	2	
13th ,,	59	90			23	55	31	38	16	7	
15th–17th ,,	——	——			=	=	——	——	——	——	
18th ,,	10	nil			6	6	nil	nil			
19th ,,	3	nil			5	11	8	nil	7	nil	
20th ,,	5	nil			10	10	2	nil	9	nil	
21st ,,	2	nil			26	32	3	nil	nil	nil	
22nd ,,	10	nil			14	12	27	nil	6	nil	
23rd ,,	7	nil			12	14	19	nil			
24th ,,	5	nil			14	10	7	nil			
25th ,,	9	nil	33	105	7	12	7	nil			
26th ,,	12	nil	34	90	7	9	14	nil	325*	nil	
27th ,,	5	nil	25	95	4	8	14	nil	52	nil	
28th ,,	9	nil	21	105	13	7	9	3	24	nil	
29th ,,	3	nil	16	123	6	13	18	nil	28	nil	
30th ,,	5	nil	10	126	10	18	10	1	63	nil	321
			——	——							
31st ,,	15	nil	16	nil			3	nil	185*	nil	231
1st September	6	nil	7	nil	36	32	13	nil	29	nil	275
					——	——					
3rd ,,	5	1	1	nil	8	nil	14	nil	32	nil	142
5th ,,	7	nil	6	nil	10	nil	14	nil	74	nil	261
6th ,,	15	nil					9		158*	nil	321
9th ,,									26	nil	60
12th ,,	22	nil	6	nil	1	nil	4		180*	nil	83
18th ,,	9	nil	2	nil	7	nil			31	nil	35
19th ,,	11	nil	4	nil	2	nil			4	nil	41
24th ,,	12	nil	4	nil							
25th ,,	9	nil	5	nil	16	nil	15	1	46	nil	31
27th ,,	7	nil			17	1	6	nil	15	nil	62
29th ,,	9	2			3	nil	11	nil	221*	nil	32
1st October	2	nil	2	nil							23
6th ,,	8	1	nil	nil	1	nil					
9th ,,							3	nil	12	nil	25
14th ,,	nil	nil			nil	nil		nil			
15th ,,	nil	nil	nil	nil	nil	nil	nil	nil			4
22nd ,,	1	nil			nil	nil	nil	nil	5	nil	
23rd ,,	nil	nil			1	nil	1	nil			

—— marks inside spraying of rooms.

= = marks outside barrier treatment.

Other explanations are given in the text.

Test Group A.

This group consisted of three adjoining houses, each containing three rooms, and one house some 50 yards distant, all in the highest part of the village. In each of the three adjoining houses, one of the rooms was sprayed with DDT, and the other two were left as controls. In the single house one ablution room—showing a high concentration of sandflies—was treated, and comparison counts made in three other rooms. Figures for test rooms represent, therefore, total counts of four rooms and for controls of nine rooms. After spraying (15th August), counts in treated rooms dropped to nil and remained nil or very low until 6th October. After that date (and in the other experiments, also, after the 6th–9th October), counts in the untreated control rooms fell to nil or to very small numbers, so that no significant comparison was possible any more. This early close of the sandfly season restricted the observation period on residual DDT effect in test group A to 52 days.

During this period, the count in the treated rooms was only a single sandfly on two occasions and two on another (see Table II); the one individual seen and caught on 6th October was dying.

Neither in this nor in the other groups was there an indication of a difference in efficiency between the two different dosages and spraying procedures used, the few and low positive counts made in treated rooms being distributed between rooms treated in one or the other way. Results in all treated rooms are therefore tabulated together.

An interesting phenomenon observed in this group is the general reduction of sandfly density in untreated rooms after spraying in the test rooms. In the three adjoining houses test and control rooms were directly contiguous and doors mostly left open or replaced by curtains. It appears from the Table that after spraying of the test rooms, sandflies were always observed in the " controls " throughout the observation period, but the average daily count was about six times lower than before spraying. This reduction of density in untreated sections of buildings or compounds partially treated with DDT can only be explained by the combination of two factors; considerable movement of sandflies between rooms, and the low rate of replacement of adults due to the long life-cycle with a larval stage lasting six weeks and more.

Test Group B.

This consisted of three rooms, forming the ground-floor of a house near group A in the higher part of the village. Rooms had plaster ceilings which greatly facilitated the counting of sandflies.

This very suitable test house was only discovered while the experiment in group A was already under way. Figures under " test rooms " are for one room, and under " controls " for the two remaining, adjoining, rooms. The high pre-treatment counts show the still considerable sandfly density in the upper village at that time, and prove that the decline of counts in the untreated rooms of group A nearby was due to the above-described " secondary effect " of DDT treatment in adjoining rooms and not to a seasonal decrease of sandflies.

Spraying was carried out on the morning of 31st August and observation time on residual effect in this group was thus limited to 36 days (until 6th October). Dosage in the single treated room was 100 mg. DDT/sq. ft. and the centre of the ceiling was left untreated. Protection was complete throughout the 36 days.

Dead sandflies were collected on sheets of paper laid out in the treated room on several occasions and as late as 6th October.

The " secondary DDT effect " in the untreated rooms is also evident in this group.

Test Group C.

This consisted of an isolated two-storied house, its courtyard and out-buildings, situated halfway down the village. The ground-floor is occupied by a cattle byre and stores, the upper storey by the living quarters, kitchen, etc.

Here, a different method for the protection of the inhabitants was tried out. Rooms inside were left untreated but the outside walls of the house were sprayed with DDT at a dosage of 2 g./sq. metre, up to 3 metres from the ground; all vertical surfaces within 50 metres of the house (stone walls, masonry of a well, etc.) and the cattle byre in the ground floor were also sprayed.

The purpose of this method was to create lethal barriers between the breeding places and the living rooms (taking into account their hopping manner of flight in short stages) and also to contaminate the immediate vicinity of the breeding places in order to kill the sandflies when emerging or returning for oviposition.

In theory, this method should have two advantages over inside treatment :—

Inhabitants would be protected not only in their rooms but also when sitting or standing outside in the evening; and the continuous killing of sandflies near their breeding places should reduce considerably, or possibly eradicate, the local fly population.

In Table II, under Group C, the broken lines below 13th August mark the spraying of outside surfaces as described above (on 15th August). Figures under "controls" are the total counts of the 3 front rooms and under "test rooms" of the 5 back rooms in the upper storey. Both front and back rooms were left *untreated* until 1st September. By this time it had become clear that the desired protection of the rooms had not been achieved by the outside barrier treatment. There was only a slight reduction of total counts during the first few days following outside spraying and by 1st September counts had reached pre-treatment levels.

It was, therefore, decided to spray the 5 back rooms inside on 2nd September. After this treatment counts fell to nil in the treated rooms and remained so—with the exception of one single sandfly on 17th September—until the end of the observation period (6th October), i.e. for 32 days.

Rooms in this house also had plaster-ceilings which facilitated the counting.

While the outside barrier treatment failed to give satisfactory protection of the rooms in the house, two effects of the treatment could be observed :—

1. Three nights prior to the spraying 4–10 sandflies were collected on outside walls but none was found again during the whole observation period afterwards on the sprayed walls.

2. People sitting outside on the porch at nights in the period before the spraying had been viciously bitten. After spraying, biting ceased completely. These observations seem to indicate that sandflies in the rooms of the upper storey had got there from distances over 50 metres, and that they had flown over these distances and up to the windows of the second storey without resting on the DDT-treated barriers in between, or on the lower (also treated) part of the house wall.

Test Group D.

This consisted of a small house and two detached latrines in a courtyard approximately 200 metres below Group C. One room in the house was treated with DDT (2 g. per m^2) the two other rooms and the two latrines were used as "controls". Spraying was carried out on 17th August.

The sprayed room was efficiently protected up to the end of the observation period (9th October) of 52 days. On three occasions only 1–3 sandflies were counted in it. (See Table II.)

There was no appreciable "secondary DDT effect" in the untreated (control) rooms of this group, since most of them were widely separated from the test room (in distinction to groups A and B).

Test Group E.

P. chinensis was the prevalent species in the locality where a two-storey house of fairly recent construction with plaster ceilings, was selected in the lowest part of the village.

Figures under " test rooms " represent counts in one DDT-treated bedroom in the upper storey, those under " controls " the total counts of the remaining three rooms in the upper storey and three rooms on the ground-floor.

The bedroom was sprayed on 15th August and thereafter protection was complete throughout the 58 days' observation period (until 9th October).

The counting of " controls " afforded some interesting observations in this group. Some very high figures, marked by an asterisk in Table II, represent evening counts, with the vast majority of sandflies counted in one single room on the ground-floor which, serving together with the porch as a café, was always wide open until late at night. The high counts were made whenever a member of the counting team happened to be on the spot at exactly the time when the night wind subsided. This occurred at varying hours between 9 p.m. and midnight but the phenomenon observed was always the same. Within a few minutes of the cessation of the wind, some dozens of sandflies appeared on the outside walls and soon after the high numbers recorded (up to 300) were found inside, where they were attracted by a strong light on the ceiling. No more sandflies could be found on the walls outside, 15–20 minutes later, and also the majority inside had already disappeared. These night swarms were almost entirely composed of *P. chinensis* (readily recognised by their small size and dark colour) and had little influence on the counts inside the rooms of the upper storey, where in untreated rooms rarely more than 4–5 sandflies were found, mostly *P. papatasii*, *P. chinensis* and *P. major* together.

Test Group F.

The figures in this column of Table II represent total sandfly counts of 3 rooms and a half-open verandah in a house of recent construction in the vicinity of Group E in the lower village. No DDT treatment was carried out here. The smoothly finished plaster walls and ceilings made observations easy and these counts were made so as to give additional data on the seasonal trend in density. Here also the 9th October marks the end of the sandfly season.

Discussion of Results.

The long columns of " nil " counts for rooms sprayed with DDT show, at a glance, the general efficiency of this method. In the few instances where only small numbers of sandflies were found in sprayed rooms, it is desirable to assess the partial protection still given by the DDT.

In doing this, account must be taken of the great fluctuation in daily counts under natural conditions. It is considered that significant figures are best obtained by comparing the *average* count of the 4–5 days' period, including the " breakdown " day in the treated room(s) with the average count during the same period in " controls " of the same test group, and with the five-day average count of the test room(s) before treatment.

This method of calculation shows that the reduction of sandflies in DDT-treated rooms at periods of partial breakdown is, in all test groups, still over 97 per cent. compared with pre-treatment counts, and over 92 per cent. compared with control counts.

It appears, therefore, that practically full protection was obtained in rooms treated inside by DDT residual spray for maximum observation periods of 52–58 days. Inhabitants of rooms thus treated enjoyed complete relief from bites after spraying.

As mentioned above, counts in untreated rooms of all groups as well as in other parts of the village fell to very low figures after 6th–9th October. This rather early termination of the sandfly season limited the maximum observation period on the residual DDT effect in the experiments to 58 days.

Though this is a considerable gain over the 24 days' period in the 1944 tests recorded by Hertig & Fisher (1945), it is unfortunate that observations could not be continued since, as the figures show, the DDT was still fully effective when the season ended and the maximum duration of the residual effect could, therefore, not be established.

Protection afforded by DDT residual spray was equally satisfactory against all three species, *P. papatasii*, *P. major* and *P. chinensis*.

The observation that sandflies decrease in numbers in untreated rooms of houses, the other rooms of which have been treated, is considered to be of great practical importance. It was observed that the greater the proportion of treated to untreated rooms, the more pronounced was the reduction. It seems justified to assume that a practical control scheme in which no rooms are left untreated and such sandfly shelters as latrines, stables, stores, etc., are sprayed would, by the continuous killing of the adults, bring about a very marked reduction of the total local population. The effect would be still greater, if the first DDT treatment of the season was carried out just before the expected emergence of the first generation after hibernation, which in Palestine is the end of April.

The failure of the outside barrier treatment in Group C seems of less importance, considering this general effect of room spraying. The observations in this experiment showed that sandflies disappeared on the outside structures that have been sprayed so that those in the house probably came from distances outside the 50-yard radius treated. Spraying of wider areas should therefore give better results. In fact, Hertig (1945) experimenting with DDT for sandfly control in Peru by the area-barrier principle, obtained satisfactory results, in that there was a marked reduction of sandflies in untreated houses by spraying outside structures over wide areas.

At present, the inside spraying of rooms is considered the best method for practical purposes for the following reasons :—

1. Immediate and complete protection of rooms, irrespective of prevailing sandfly density, has been conclusively proved.

2. The amount of spraying solution (and DDT) for outside barrier-spraying in a sufficiently wide area is much higher than that required for inside treatment.

3. As explained above, the observations on the " secondary DDT effect " in untreated rooms justify the assumption that the density in general will be greatly reduced in a locality where a large scale inside treatment of houses with DDT is carried out.

Influence of DDT Treatment on Sandfly Fever Incidence.

During the summer of 1945 all military camps and installations in Palestine were, as a routine measure, sprayed regularly (approx. every two months) with DDT. Spraying began generally at the end of April, with some delay in a few locations.

In appreciation of the effect of this measure on the incidence of sandfly fever, the complicating factors that have to be taken into consideration are percentage of non-immune newcomers, and correctness of diagnosis and accuracy of reported numbers.

As there is no clinical sign or laboratory test by which a fever of short duration can, with absolute certainty, be diagnosed as sandfly fever, inaccuracies in the numbers reported are unavoidable.

The proportion of susceptible personnel can also be only estimated—within wide limits—for technical reasons.

No figures are, therefore, given to show the decrease in sandfly fever cases, since these figures would be too inaccurate for statistical conclusions.

The following general statement, however, can be made. While non-immune replacement troops in Palestine were rather more numerous in 1945 than in 1944, and the same sandfly-infested areas were occupied in both years, the sandfly-fever rate in 1945 was much lower (approximately one-tenth) than in 1944. There was also a very marked reduction in cases of P.U.O. (Pyrexia of unknown origin) in the summer of 1945 as compared with 1944. The latter fact is important as a proportion of P.U.O. cases are really sandfly fever cases.

Fortunately there was one locality in which the observations of the influence of DDT on sandfly-fever incidence can be regarded as conclusive.*

The locality in question was an R.A.F. station known throughout the war for the abundance of its sandflies and cases of sandfly fever.

As the station was connected with a training establishment, there was a continuous supply of non-immune personnel in 1945 and during the previous years. Special attention was paid by the medical officer in charge to the diagnosis of sandfly fever.

Heavy infestation was noted in this station in spring 1945, and large numbers of fever cases began to appear in May to early June. Large scale DDT treatment was then carried out (mid-June) followed by six-weekly re-applications until October.

Sandfly fever incidence fell abruptly at the end of June, and remained negligible throughout the ordinary peak-season (August–October).

Summary.

Field experiments carried out in 1945 at Rosh Pina in Palestine have proved the great efficiency of DDT residual spray in houses against sandflies.

A high degree of protection against *P. papatasii*, *P. major* and *P. chinensis* was obtained by inside spraying of rooms with DDT in kerosene.

The residual action lasted for 52–58 days during the hot summer months. At the end of this period the effect was still undiminished but the end of the sandfly season made further observations impossible. Judging from observations made elsewhere it may be assumed that the residual effect would have lasted not more than another two weeks.

Dosage of 1 g. per m^2 (100 mg. DDT per sq. ft.) was fully effective, and 2 g. per m^2 gave no better results. The best method of application is the treatment of all internal surfaces (walls and ceiling). As sandflies tend to congregate in the angle between wall and ceiling, treatment of walls and this angle only gives equally good results.

An attempt to protect a house by spraying " barriers " (vertical surfaces of stone walls etc., within a radius of 50 metres) failed. Sandflies on the outside surfaces disappeared after the DDT treatment but the observations recorded here and those reported by other authors suggest that much larger areas would have to be sprayed in order to make this method effective.

A marked reduction of sandflies in *untreated* rooms followed the spraying of other rooms in the same house with DDT. The mechanism of this phenomenon is explained. A *general reduction* of the sandfly population can be expected when large-scale DDT treatment is carried out in a village or camp area.

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* I am indebted to Sq.-Ldr. E. Russel for this information.

Some observations on the influence of DDT treatment on sandfly fever incidence in military camps are recorded.

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TOXICITY OF DDT APPLIED TO LIMEWASH.

By A. Hadaway and F. Barlow,
Colonial Insecticide Research (Uganda).

ge.

Reports on the effectiveness of DDT applied to the limewashed interiors of buildings have been conflicting. This question is of importance in the tropics where so many European houses are limewashed. Laboratory tests recorded here were made to determine, firstly, how effective DDT was when applied to limewash, and, secondly, the cause of any loss of toxicity.

Methods.

Limewash made from partly slaked lime supplied by P.W.D., Entebbe, was applied with a brush to glass plates 6 inches square but apparatus for the application of a known dosage to a given surface area was not available. The limewashed glass plates together with clean " pilot " plates were, therefore, placed on the ground in a measured area which was sprayed with a known volume of insecticide from a Four Oaks " Kent " Sprayer. Deposits obtained by this method varied considerably over the treated area. The glass plates were then kept in the laboratory.—

The analysis of the lime used in the tests was as follows :

Insoluble in 10 per cent. hydrochloric acid	8·6 per cent.
Soluble in 10 per cent. hydrochloric acid :—	
Calcium oxide, CaO	39·8 per cent.
Calcium carbonate, $CaCO_3$	36·6 per cent.
Combined water	8·0 per cent.
Ferric oxide, Fe_2O_3	2·6 per cent.

Notes : 1. Part of the lime was unslaked.

2. Some alumina and magnesium were present but not estimated.

3. The iron content is high but from chemical estimations of the insecticide it does not seem that any marked decomposition of the DDT was produced.

4. Qualitative tests showed chlorides and sulphates to be absent.

5. The alkalinity of 1 g. of lime was equivalent to 14·22 ml. of normal acid when titrated with phenolphthalein as indicator.

The test insect, *Glossina palpalis*, R.-D., was bred in the laboratory from pupae collected on the lake shore. All specimens were given the opportunity to feed on sheep daily. Flies seven to ten days old were exposed to the treated surface individually in 3×1 inch tubes, which were then kept in racks over water in special trays. Mortality counts were made 24 and 48 hours after contact. The sexes of the flies are not given in the tables as it has been found that there is no significant difference in the resistance of the male and female flies.

Control deaths were negligible and are not given in the tables ; of the first 1,000 flies used as control, 8 only died in 24 hours and 35 in 48 hours.

In some tests mosquitos have been used ; they were exposed individually in tubes in exactly the same way. *Aëdes aegypti*, L., were laboratory bred, 2–4 days old, and fed daily on guinea pigs ; *Anopheles gambiae*, Giles, were fully-fed " wild " specimens collected from native huts early in the morning of the day of the test.

The method adopted for chemical estimations of deposits was dehydrohalogenation followed by titration of the liberated chlorine ions with silver nitrate solution. The insecticide was first extracted from the removed layer of limewash by shaking with benzene and filtering through cotton wool. Blanks were carried out on untreated limewash.

Limewash treated with DDT/Kerosene Solution.

(a) *Preliminary test with limewashed plaster.*

Blocks of limewashed cement and plaster were cut from old buildings. One block had been limewashed six months before, one nine months, and one 18 months before; the layers of limewash were about 1 mm. thick. They were sprayed with 5 per cent. DDT (83 per cent. pp′) in power kerosene and *G. palpalis* were exposed to them 48 hours later for 20 seconds. Results, given in Table I, show that no kills were obtained although the DDT recovered varied from 114 to 192 mgms. per sq. ft.

TABLE I.

Age of block	Percent. kill in 24 hours	DDT deposit recovered
6 months	5	162 mgms. per sq. ft.
9 ,,	0	192 ,, ,, ,, ,,
19 ,,	0	114 ,, ,, ,, ,,

(b) *Fresh limewash.*

Five per cent. DDT (83 per cent. pp′) in power kerosene was sprayed on a limewashed glass plate 3 hours after application of the lime. *G. palpalis* were exposed to this for 20 seconds 24 and 48 hours later. Percentage mortalities (Table II) were very low compared with those obtained on the "pilot" glass plate treated with DDT only.

TABLE II.

Age of DDT deposit	Treatment of glass plate	No. of flies	Percent. kill in 24 hours	Percent. kill in 48 hours	DDT recovered mgms./sq. ft.
24 hours	Limewash only ...	30	0	0	0
	Limewash and DDT ...	30	10	16·7	236
	DDT only	30	86·7	90	173
48 hours	Limewash and DDT ...	30	6·7	23·3	
	DDT only	30	96·7	96·7	

(c) *Limewash of different ages.*

Glass plates were limewashed and kept in the laboratory for varying periods from 3 hours to 8 weeks before treatment with 5 per cent. DDT (83 per cent. pp′) in power kerosene. Flies were then exposed for 20 seconds to the treated surfaces 24 hours after the application of insecticide with the results shown in Table III. Loss of toxicity not only occurs when the insecticide is applied to fresh limewash, but also to limewash two months old.

The weight of lime on these plates varied from 7·6 to 13·6 gms. per sq. ft.

TABLE III.

Treatment of glass plate	Age of limewash when treated with DDT	No. of flies	Percent. kill in 24 hours	Percent. kill in 48 hours	Recovery of DDT in mgms./sq. ft.
Lime and DDT ... DDT only	3 hours —	30 30	23·3 100	33·3 100	64 106
Lime and DDT ... DDT only	1 day —	20 20	10 100	35 100	85·9 89
Lime and DDT ... DDT only	4 days —	30 30	46·7 100	46·7 100	176·3 104
Lime and DDT ... DDT only	7 days —	30 30	23·3 100	30 100	150·4 144
Lime and DDT ... DDT only	9 days —	30 30	20 93·3	20 93·3	171·5 104·5
Lime and DDT ... DDT only	3 weeks —	30 30	26·7 100	26·7 100	312 142·4
Lime and DDT ... DDT only	8 weeks —	20 20	35 100	45 100	240 152

36 9·

(d) *Adsorption.*

Tests were conducted to decide whether the loss of toxicity on lime was due to adsorption of DDT on the lime particles.

A weighed quantity of DDT (pure) was dissolved in 20 ml. power kerosene. 10 gms. of the lime used in the toxicity experiments were added and the mixture was shaken occasionally for about half an hour. The lime was filtered off through cotton wool, a sample of the filtrate taken and the DDT content determined by the dehydrohalogenation method. A blank was carried out on an equal unhydrolysed sample.

In this way the amount of the DDT removed from solution by 10 gms. of lime was found. The lime was not homogeneous as shown by the varying blank estimations. In the range of concentration of DDT used the amount of insecticide adsorbed is independent of the concentration. However, the experiment sufficed to show that the amount of DDT inactivated on sprayed limewash by adsorption is very small and does not explain the non-availability of the insecticide. DDT recovered after shaking in power kerosene with 10 gms. lime :—

	No. of Sample			
	1	2	3	4
Mgms. DDT added	2·6	4·3	6·4	8·1
Mgms. DDT recovered	1·9	3·5	4·7	7·4

The weight per sq. ft. of the lime layer on the glass plates in tests (c) varied from 7·6 to 13·6 gms. In subsequent tests thicker layers weighing up to 80 gms. were used. Even in determination 3, if 10 gms. of lime adsorbed 1·7 mgms. DDT, 80 gms. of lime would only adsorb about 14 mgms. DDT out of the amount sprayed.

(e) In several instances the part of a benzene extract from treated lime not used for analysis was evaporated to dryness and the residue crystallised from ethyl alcohol.

In this way from 30–60 per cent. of the DDT found present by analysis was recovered and identified as pp′ DDT by the melting point and mixed melting point compared with those of an authentic sample. As the crystallisation leaves some

DDT in the mother liquor it can be assumed that a large proportion of the DDT applied has not lost its toxicity because of decomposition.

Extraction was made in this way from glass plates which had been limewashed and treated with DDT and which subsequently showed little or no toxicity to flies exposed for 20 seconds. 20 mgms. of the crystallised extract were dissolved in ether and applied to a clean glass plate to give a dosage of 80 mgms. per sq. ft. *G. palpalis* were exposed to this three days later for 20 seconds and a kill of 83·3 per cent. was obtained in 24 hours.

(f) *Longer exposures to treated lime.*

A glass plate limewashed 8 weeks previously was sprayed with a 5 per cent. solution of DDT in power kerosene. One week later *G. palpalis* were exposed to the treated surface for various periods ; kills are given in Table IV. From the limewashed plate 240 mgms. DDT per sq. ft. were then recovered and 152 mgms. from the plates treated with DDT only.

TABLE IV.

Treatment of glass plate	Exposure time	No. of flies	Percent. kill in 24 hours	Percent. kill in 48 hours
Lime and DDT	20 secs.	20	25	40
„ „ „	1 min.	20	60	65
„ „ „	2 min.	20	60	90
„ „ „	5 min.	35	80	89
DDT only	20 sec.	30	100	100
Controls	—	20	0	0

A glass plate limewashed one week previously was sprayed with a 5 per cent. DDT solution in power kerosene. Flies and mosquitos were exposed to the treated surface 24 hours, 2 weeks and 4 weeks after the application of insecticide. Percentage mortalities are given in Table V.

At the end of four weeks 378 mgms. DDT per sq. ft. were recovered from the plate ; the weight of the lime layer was 85 gms. per sq. ft.

TABLE V.

Percentage mortalities 24 hours after contact with treated limewash on a glass plate. Numbers of insects used are given in brackets.

Species	Exposure time	Time after application of Insecticide.		
		24 hours	2 weeks	4 weeks
G. palpalis	20 sec.	5 (20)	—	—
	2 min.	30 (20)	50 (20)	40 (20)
	5 „	55 (20)	60 (20)	70 (20)
	10 „	80 (20)	75 (20)	80 (20)
	15 „	96 (25)	— —	—
Aëdes aegypti	1 hour	49 (35)	40 (62)	37 (77)
	2 hours	82 (35)	82 (80)	73 (40)
Anopheles gambiae ...	½ hour	66 (35)	60 (30)	—
	1 „	97 (35)	83 (34)	—

Flies and mosquitos were exposed concurrently to 50 mgms. DDT per sq. ft. on a clean glass plate. Percentage mortalities are given in Table VI.

TABLE VI.

Percentage mortalities 24 hours after contact with 50 mgms. DDT per sq. ft. Numbers of insects used are given in brackets.

Species	Exposure time	Time after application of Insecticide.		
		24 hours	2 weeks	4 weeks
G. palpalis	5 sec.	92 (60)	77 (40)	77 (35)
	15 ,,	100 (60)	85 (40)	87 (30)
Aëdes aegypti ...	2 min.	66 (105)	38 (32)	—
	5 ,,	95 (112)	55 (40)	38 (40)
Anopheles gambiae ...	½ min.	65 (60)	—	—
	1 ,,	71 (95)	32 (25)	30 (20)
	2 min.	100 (60)	63 (30)	50 (45)

It seems clear that loss of toxicity of DDT applied to limewash as a kerosene solution is not due to chemical change or decomposition for most of the insecticide can be recovered. The amount of DDT adsorbed by the lime particles is very small and does not explain the non-availability of the insecticide.

The loss of toxicity is due mainly to absorption and the consequent masking of the insecticide. Insects must remain in contact with the treated limewash for much longer periods than is necessary on simple DDT deposits to obtain similar results.

In these tests the layer of lime on the non-absorbent glass has been only a very thin one, much thinner than that applied in practice to the plaster or cement walls of buildings. That toxicity is lost on the limewashed glass plates where absorption cannot be very deep suggests that the available insecticidal layer is a very thin one.

Addition of an Adhesive to the Limewash.

Two glass plates were limewashed as previously described whilst two other plates were treated with the limewash used by the P.W.D. for European houses. The latter contains 68 lb. lime, 2 lb. glue, 2 lb. muwogo flour (Cassava) and ½ lb. salt.

TABLE VII.

Treatment	Age of DDT deposit	No. of flies	Percent. kill in 24 hrs.	Percent. kill in 48 hrs.	DDT mgms. per sq. ft. re-covered	Wt. of lime gms./sq. ft.
Lime and Gum and DDT (1)	24 hours	25	32	40	256	40
	2 weeks	20	30	45		
Lime and Gum and DDT (2)	24 hours	25	40	48	556	47
	2 weeks	20	30	45		
Lime and DDT (1) ...	24 hours	25	40	52	300	67
	2 weeks	20	45	50		
Lime and DDT (2) ...	24 hours	25	48	56	296	60
	2 weeks	20	50	60		
DDT only	24 hours	25	100	—	152	0
	2 weeks	20	100	—		

The layer of lime was much thicker than that used in previous tests; deposits varied between 40 and 67 gms. per sq. ft.

When the plates were dry they were sprayed heavily with a 5 per cent. DDT solution in power kerosene. *G. palpalis* were exposed to them for 20 seconds, 24 hours and 2 weeks after the application of insecticide. Percentage kills are given in Table VII.

The addition of adhesives to limewash had no apparent effect on the absorption of the insecticidal solution.

Limewash treated with an Emulsion.

Glass plates limewashed one week previously were sprayed with 5 per cent. DDT emulsion made by adding 4 parts of water to 1 part of A.19 (T.P.543) concentrate.

G. palpalis were exposed to them for 20 seconds 24 hours and 2 weeks after the application of insecticide. Results are given in Table VIII.

A loss of toxicity also occurs when an emulsion is sprayed on to lime.

TABLE VIII.

Treatment	Age of DDT deposit	No. of flies	Percent. kill in 24 hrs.	Percent. kill in 48 hrs.	DDT mgms. per sq. ft. recovered	Wt. of lime gms. sq. ft.
Lime and emulsion (1)...	24 hours	25	44	44	154	70
	2 weeks	20	30	30		
Lime and emulsion (2)...	24 hours	25	40	48	261	80
	2 weeks	20	20	30		
Emulsion only	24 hours	25	100	—	239	0
	2 weeks	20	10	—		

Limewash treated with a Dispersible Powder.

Glass plates limewashed one week previously were sprayed with a 2 per cent. DDT suspension in water made from DDT dispersible powder Guesarol. *G. palpalis* were exposed to them for 2 minutes 24 hours and 2 weeks after the application of insecticide. Percentage kills are shown in Table IX.

Kills obtained with the dispersible powder were approximately the same on the limewashed plates as on the "pilot" glass plates. The deposit appears to remain on the surface of the lime. Longer contact times are necessary to produce reasonable kills even on the pilot glass plate. This is probably because the original Guesarol contains only 5 per cent. DDT and the large amount of inert material making up the bulk of the dispersible powder masks the effect of the insecticide.

TABLE IX.

Treatment	Age of DDT deposit	No. of flies	Percent. kill in 24 hrs.	Percent. kill in 48 hrs.	DDT mgms. per sq. ft. recovered	Wt. of lime gms. sq. ft.
Lime and Guesarol (1)...	24 hours	20	25	45	116	70
	2 weeks	20	30	40		
Lime and Guesarol (2)...	24 hours	20	50	65	189	77
	2 weeks	20	50	70		
Guesarol only	24 hours	20	35	45	120	0
	2 weeks	20	40	45		

Water Paint containing DDT.

A water paint manufactured by Walpamur Co. Ltd., light stone in colour and containing DDT (Geigy) was thinned with water and applied by brush to glass plates.

One glass plate received only a thin coating of paint. *G. palpalis* were exposed to this for 5 minutes and 15 minutes one week after the application. Percentage kills 24 hours after contact were 10 and 50 per cent. respectively. Percentage mortality of 82 *A. gambiae* exposed for 1 hour contact was 57 per cent. 70 mgms. DDT per sq. ft. were recovered from the glass plate.

Another glass plate was given a thick coating of paint. Percentage kill of *G. palpalis* exposed for 5 minutes was 13 per cent.; for 15 minutes 60 per cent. After contact of 1 hour 50 per cent. of 58 *A. gambiae* were dead in 24 hours. Subsequent chemical determination showed a dosage of 623 mgms. DDT per sq. ft.

The kills obtained with both plates are very similar. When the paint dries a large proportion of the insecticide must be hidden below the surface, and the layer of contact-available insecticide only very thin.

Conclusions.

(i) There is a marked loss of toxicity when DDT is applied to limewash as a solution in kerosene. This loss occurs not only on fresh limewash but also on limewash several months old.

(ii) The amount of DDT adsorbed by the lime particles is too small to account for the non-availability of the insecticide.

(iii) The loss of toxicity is not due to any chemical change or decomposition; most of the insecticide can be recovered. It seems that loss of toxicity is due to absorption and the consequent masking of the insecticide.

(iv) The addition of adhesives to the limewash does not prevent absorption.

(v) A similar loss of toxicity occurs when an emulsion A.19 is sprayed on to limewash.

(vi) Guesarol dispersible powder is slightly more promising. Dispersible powders with higher DDT contents are needed for trials; the large amount of inert material in Guesarol powder seems to mask the effect of the insecticide.

COCCIDS ATTACKING CACAO (*THEOBROMA CACAO*, L.), IN WEST AFRICA, WITH DESCRIPTIONS OF FIVE NEW SPECIES.

By A. H. Strickland,
Entomologist, West African Cacao Research Institute, Gold Coast.

E.H.iv.

Introduction.

Cacao was introduced into West Africa in the latter years of the last century, and for a time remained relatively free from pests and diseases. It is, in fact, only recently that two major scourges have developed to a point where they are threatening the future of the cacao industry in the Gold Coast and Nigeria. The first, the cacao Mirids (*Sahlbergella singularis*, Hagl., and *Distantiella theobroma*, Dist.) is outside the range of the present paper. The second, and most important, is a virus disease, known as "Swollen Shoot", which is transmitted from tree to tree by at least three species of mealybugs of the family Pseudococcidae.

33 65
34 182

Reference to the note of Cotterell (1943) and Box (1945), made it clear that the situation with regard to the cacao-feeding Coccids, and especially with those of the Pseudococcine series, would have to be investigated fully, both with regard to their biology and their distribution throughout the equatorial forest belt that forms the main West African cacao growing area.

The 44 species detailed in the present paper are the result of a year's intensive collecting by the writer in the Eastern Province of the Gold Coast, during which time earlier collections made by W. H. Patterson and G. S. Cotterell (late Senior Entomologists, Gold Coast Department of Agriculture) and H. E. Box (late Senior Entomologist, West African Cacao Research Institute) have been examined and worked on. In all, over 800 collections of material from cacao and indigenous and introduced plants have been examined, principally from the Gold Coast. It is probable that more species will be discovered in the future, particularly on the edges of the cacao belt, and in areas such as the French Ivory Coast, where cacao is only beginning to be grown on a large scale. The present list should not, therefore, be taken as complete.

In addition to the species taken in the field by the writer, or one of his associates, those species detailed in the literature as having been collected from cacao in west and central Africa, and on the island of San Thomé, have been included. At the end of the paper those species taken from cacao in areas other than Africa are briefly mentioned.

The Cacao Coccids.

(1). *Aonidiella gracilis* (Balachowsky, 1929).

Cited by Mayné and Ghesquière (1934) as attacking cacao in the Lusambo region of the Belgian Congo. It is common on the undersides of cacao leaves and produces a characteristic chlorosis. Also cited by Laing (1932) from Kondue (Sankuru) on the undersurface of cacao leaves.

(2). *Selenaspidus articulatus* (Morgan, 1889).

De Seabra (1920) states that this species has been observed on cacao, coffee, coconut palm, avocado pear and also on *Citrus* spp., but always rare and dispersed. He maintains that its chief food plant in San Thomé is *Lonchocarpus sericeus*.

(3). *Selenaspidus silvaticus*, Lindinger, 1909.

Cited by Mayné and Ghesquière (*l.c.*) as the species that has adapted itself to cacao, on which they collected it from the undersides of the leaves; by Lindinger (1909), from Anacardiaceae, *Rinorea exappendiculata*, and *Bandeiraea speciosa*, at Bipinde, Cameroons, from *Ficus indica* at Amani, Tanganyika; by Newstead (1917a) from *Citrus* sp. at Entebbe, Uganda, and by Laing (1932) associated with *Aonidiella gracilis*, on cacao leaves at Kondue.

(4). *Pseudaonidia trilobitiformis* (Green, 1896).

Recorded by De Seabra (1917) from cacao in San Thomé. De Seabra and Vayssière (1918) state that it is very harmful, the attack first manifesting itself at the tips of the branches, which die back little by little, until, in two or three months, the whole tree is defoliated. Cotterell has collected this species from cacao in San Thomé, and Mayné and Ghesquière cite it as occurring on cacao in the lower Congo region, where it was taken from *Citrus* sp., *Aleurites* sp., *Ficus* sp., *Ixora* and a leguminous plant of the genus *Millettia* and suggest that it has been introduced on to the mainland from San Thomé. Newstead (1911) has recorded it from oleander at Usagara, East Africa, and Laing (1928) from *Coffea* sp., *Asystasia* sp., *Ficus repens*, *Passiflora quadrangularis* and oleander from San Thomé. Hargreaves (1937) cites it from *Citrus* sp., *Coffea* sp., *Psidium guajava*, and *Nerium oleander* in Sierra Leone. It is clearly a widespread insect, with no particular host preferences.

(5). *Hemiberlesia palmae* (Morgan & Cockerell, 1893).

Recorded as "very rare" on the leaves of cacao, avocado pear and *Citrus* sp., but common on *Carica papaya* and *Artocarpus incisus* by De Seabra and Vayssière (*l.c.*) in San Thomé, and also cited by De Seabra (1917a) when he discusses its economic importance in San Thomé. Recorded also by Laing (1928) from cacao, coffee, *Carica papaya*, *Persea gratissima*, *Artocarpus incisus*, *Citrus* sp. and coconut from San Thomé. Vayssière (1913) records it from *Elaeis guineënsis* in the Cameroons. This species, originally described from coconut palms from Kingston, Jamaica, would seem to be widely distributed in West Africa.

(6). *Mytilococcus beckii* (Newman, 1869).

Cited by De Seabra (1919) from cacao in San Thomé, and subsequently (1919a) from *Persea gratissima*. The writer has taken this species commonly from *Citrus* spp. in the Gold Coast.

(7). *Pseudaulacaspis pentagona* (Targioni-Tozzetti, 1885).

Recorded by De Seabra (1920) from cacao, coconut and *Loranthus* sp., in San Thomé who states that this species would probably be an important cacao pest were it not controlled by an entomophagous fungus, *Microcera cocophila*. Cited also by De Seabra and Vayssière (*l.c.*) from papaw and *Hevea* rubber, also from San Thomé.

(8). *Ceroplastes destructor*, Newstead, 1917.

The earlier records of this species are listed as *C. ceriferus*, Anderson, and it was not until 1917 that Newstead described it as a distinct species. Newstead (1910a) stated Gowdey had recorded it from coffee in Uganda in sufficiently large numbers to be of economic importance, and also from cacao, *Agave*, *Canna*, *Croton* and *Hibiscus* sp. Newstead (1911) also cites it from *Coffea arabica* at Magrotto, in East Africa, and Gowdey (1913) from guava in Uganda.

(9). *Ceroplastes bussei*, Newstead, 1906.

This species was recorded by von Faber (1909) from cacao in the Cameroons. It was named by Newstead in 1906, citing cacao as the host, and Cameroons as the locality but no description was given. As a description, subsequently submitted for publication, was destroyed by fire before publication, the name is a "nomen nudum" and should be ignored in future.

(10). *Ceroplastes lamborni*, Newstead, 1917.

Described from a collection from cacao and "a large climber on a bush tree" made by Dr. W. A. Lamborn, at Ibadan, Nigeria.

(11). *Ceroplastes theobromae*, Newstead, 1908a.

No references can be found to this species in the literature apart from the original description, which quoted cacao as the host, and Soppo, Cameroons, as the locality.

(12). *Coccus elongatus* (Signoret, 1873).

There is a slide in the British Museum collection recording this species from cacao in Uganda.

(13). *Hemilecanium imbricans* (Green 1903).

This species was also described by Newstead (1908a) as *H. theobromae*, from cacao at Soppo, Cameroons. It was originally collected from cacao by Dr. Busse in 1905 and is cited by von Faber (*l.c.*) from cacao in the Cameroons.

(14). *Inglisia theobromae*, Newstead, 1917.

Described from material collected by Gowdey "on stems of cacao pods and flowers" from Nagunga, Uganda.

(15). *Inglisia conchiformis*, Newstead, 1910b.

Described from *Averrhoa carambola* from Konakry, West Africa, and *Haronga madagascariensis* from Uganda, and recorded from cacao in Uganda by Gowdey (1916). Newstead cites the larvae of *Eublemma scitula*, Ramb. as predators.

(16). *Pulvinaria aristolochiae*, Newstead, 1917.

Described from *Aristolochia* sp. at Aburi, Eastern Province, Gold Coast. Subsequently taken twice by the writer on cacao, in both cases as single adult female specimens, once from Pimpimso (also in the Eastern Province), and once from Tafo. One of the specimens was taken just before it started to form its ovisac; it was an elongate pyramidical shape, and covered with thin flakes of a dirty yellowish wax. An extremely heavy infestation of this species developed on a mango tree at Tafo in 1944. Numerous adults were present on all of the smaller branches and twigs, which were covered with the conspicuous felted ovisacs. The mango adults were also a dirty yellowish colour and, after depositing their ovisacs, very much flattened and wrinkled.

(17). *Pulvinaria jacksoni*, Newstead, 1908.

Described from cacao at Calabar. This species is extremely common in the Gold Coast. At Tafo, its chief food plant is *Acalypha* spp., from which all stages, including the male cocoons, have been collected. It has not, however, been collected from cacao at Tafo as yet. With regard to the *Acalypha* material, the adult females, before extrusion of the ovisacs, were green in colour, with patches of brown along either side of the mid-dorsal line and on the margin. At this stage, the insects were about 6 mm. long, 4 mm. wide, and 3 mm. high, and pyramidical in shape. Extrusion of the ovisac (4 by 40 mm.) took about 4 days, during which time the insects shrunk to a little over 1 mm. in height. The young female larvae were a uniform shiny green in colour, the male larvae being brown-black, with the tip of the mid-dorsal keel white. During pupation the male cocoons became cemented to a leaf, and turned a uniform white. This species was recorded by von Faber (*l.c.*) from cacao in the Cameroons. Other African records include :—Mayné and Ghesquière (*l.c.*), from cotton at Kasai, cotton at Ibadan (Peacock, 1913), cotton in Nyasaland (Ballard, 1913), cotton at Kyerima, Kenya (Newstead, 1911), *Aranthemum bicolor* at Kampala, and granadilla at Nairobi (Newstead, 1917a), *Ficus* sp. at Dakar (Newstead, 1908), and tung oil trees in Nyasaland (Smee, 1940). Waterston (1916) described an Encyrtid parasite, *Chiloneurus afer*, which was bred out from *P. jacksoni* at Aburi by W. H. Patterson.

(18). *Saissetia subhirsuta*, Newstead, 1916.

A species of *Saissetia* that agrees extremely well with Newstead's macroscopic description of *S. subhirsuta*, but differs slightly microscopically in the size and distribution of the lateral setae, was collected once from cacao at Tafo by Box. The same species was collected in the field at Tafo by the writer, heavily infesting the growing point of a mature *Canthium glabriflorum* (Rubiaceae). Gold Coast records given in the type description are :—*Tabernaemontana* sp., *Landolphia* sp., *Oroxylon* sp., and *Garcinia* sp., all at Aburi, and (type host and locality), *Blighia sapida* at Odumasi, Eastern Province. This species has also been recorded from Sierra Leone, on *Cola nitida* (Hargreaves, 1937).

(19). *Stictococcus aliberti*, Vayssière, 1936.

In his description Vayssière states the species had been collected on cacao in Togo on the Daye plateau (around Kloutou) in January 1934. This is the only record known to the writer.

(20). *Stictococcus diversiseta*, Silvestri, 1914a.

The identity of this species, when compared with series of *S. dimorphus*, Newst. and *S. multispinosus*, Newst., is by no means clear, and it is possible that it will fall as a synonym when a larger amount of material is to hand. Specimens of what is believed to be this species were taken from *Ceiba pentandra* at Tafo and have been compared by the writer with specimens determined by Silvestri with which they agree well. They also agree well with *S. dimorphus* in many respects. The two have been tentatively separated on external appearances. The Tafo material was subcircular in shape, approximately 1·8 by 2·0 mm. (no larger specimens were noted) with the body colour a pale lemon yellow. Around the lateral margin were clumps of white wax, about the size of the head of a pin, with, dorsally, a horse-shoe shaped band of wax around the anal aperture, with the free ends projecting posteriorly. Anteriorly there were two distinct parallel bands of white wax on the dorsum, these being slightly broader posteriorly than anteriorly. There was no sign of the distinct " Broad V-shaped depression on the dorsum in front of the anal orifice " mentioned by Newstead in his description of *S. dimorphus*. *S. diversiseta* was described from cacao and *Annona* sp. from Cotonou, French Togoland and has since been recorded by Gowdey (1917) from *Annona muricata*, *Artocarpus integrifolius*, *Cajanus indicus*, *Croton tiglium*, *Dolichandrone platycalyx* and *Hibiscus* sp. from Uganda. In the same paper Gowdey states that the larvae of *Eublemma costimacula*, Saalm. (Lepidoptera, Erastriinae) are predacious on this insect. Waterston (1916a) cites an Encyrtid parasite, *Aethognathus afer*, Silv., bred from it at Aburi by W. H. Patterson, and Silvestri himself (1914) described *Tetrastichus stictococci*.

(21). *Stictococcus dimorphus*, Newstead, 1910a.

Described from cacao in Uganda, and cited from the same place by Gowdey (1913) who also records it from *Cajanus indicus*, *Croton tiglium*, *Annona muricata*, *Markhamia platycalyx* and mulberry. It has also been taken in Nigeria (Lamborn, 1914), " . . . another *Stictococcus*, *S. dimorphus*, occurred on cacao at Agege, but only lightly on native farms, probably due to pigeon pea being used as shade (on which there were very heavy infestations)." It was being held in check by the larvae of *Eublemma scitula*, Ramb. Waterston (1916a) cites an Encyrtid, *Aethognathus afer* var. *cavilabris*, Wat., as having been bred from this species at Entebbe by Gowdey. The species has not as yet been taken in the Gold Coast.

(22). *Stictococcus formicarius*, Newstead, 1910.

Described from Stanleyville, Belgian Congo, from the hollow branches of *Barteria fistulosa* and *Cuviera angolensis*, attended respectively by the ants *Sima spininoda*, Andre and *Crematogaster* (*Atopogyne*) *africana* subsp. *laurentis*, Forel and originally collected by P. H. Kohll. Mayné and Ghesquière (*l.c.*) record it as able to make lesions on cacao, coffee and *Annona muricata* in the Congo, and like all the *Stictococcus* this species is protected by *Oecophylla*, and often by species of *Crematogaster* and *Pachysima*. The same authors also cite Laing's var. *tuberculatus* (Laing, 1932) as occurring in colonies on the young branches of *Sterculia tragacantha*, large numbers of which are destroyed by a carnivorous caterpillar of the genus *Eublemma*. Gowdey (1917) records this species from *Ficus* sp. in Uganda.

(23). *Stictococcus gowdeyi*, Newstead, 1913.

Described from Entebbe, Uganda, on *Haronga madagascariensis*. Subsequently, Newstead (1917) cites it : " . . . on young shoots (of cacao), invariably attended by the ant *Oecophylla* " from Southern Nigeria. Gowdey (1917) records it from coffee in Uganda, and Mayné and Ghesquière from cacao in the Congo. Box has collected it twice on cacao in the French Ivory Coast, Cotterell once from cacao at Kwabeng (Eastern Province) and once from Tafo, and the writer once from *Brillantaisia nitens* at Tafo. The *Brillantaisia* specimens were between 2 and 3 mm. in diameter, and a deep cherry-red to red-black in colour, with a thin band of white wax fixing the yellow lateral setae to the host plant. Gowdey (1917) states that this scale is parasitised by two Eulophids, *Coccophagus comperei*, Gir., and *Epitetrastichus ugandensis*, Gir., in Uganda.

(24). *Stictococcus intermedius*, Newstead, 1917.

Described from cacao at Aburi. This is the only record known to the writer.

(25). *Stictococcus multispinosus*, Newstead, 1908c.

Box has collected this species twice from cacao in the French Cameroons, and Cotterell once in the Gold Coast. Mr. A. F. Posnette, Senior Pathologist, recently brought in a large number of specimens of this species from cacao at Wiawso, in the Western Province of the Gold Coast. He said that they were the commonest cacao Coccids in the area at the time of collection. The writer has taken this species from *Piper umbellatum* at Tafo. These specimens were subcircular in shape, approximately 1·8 by 1·8 mm., the body colour being a light to dark brown, with a marginal band of white wax of a lumpy consistency. On the dorsum were four triangular wax plates, giving the insects the appearance of " hot cross buns". The body surface beneath the wax covering was a dull matt red to brown, with many short, light brown setae dorsally and laterally. First-instar larvae were minute, oval and a bright red in colour. The second stage nymphs were lighter in colour than the adults, with the marginal wax band, and two pairs of dorsal and dorso-lateral wax bands, each tapering to a blunt point at either end. Other African records are :—Newstead (1917) from pigeon pea stems at Agege, Southern Nigeria, and from *Markhamia platycalyx* in Uganda, and (1917a) from *Cola* sp. at Aburi. Hargreaves (*l.c.*) records it from *Cassia* sp. and *Cola nitida* in Sierra Leone.

(26). *Stictococcus sjostedti*, Cockerell, 1903.

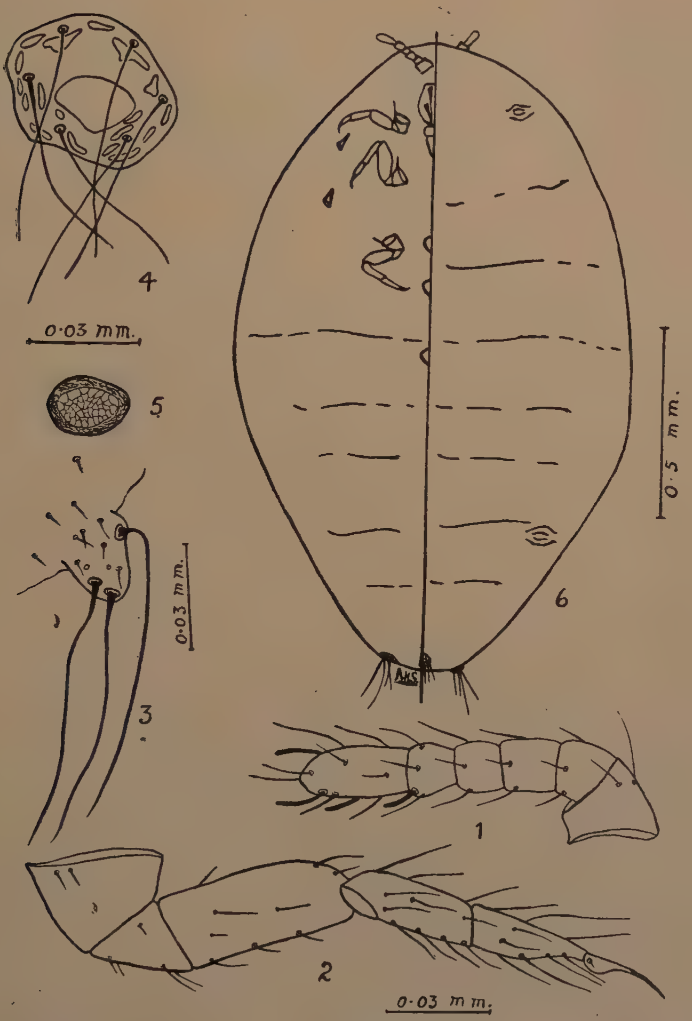
This species is easily the commonest scale insect of Gold Coast cacao and its early history is of some interest. After Cockerell described it, Newstead (1908), received some material for his collection from Dr. Winkler collected in 1904 from cacao pods at Victoria, Cameroons. Subsequently, Dr. Slater Jackson collected specimens from cacao and rubber at Gaboon, cacao at Aburi and coffee at Warri. Von Faber (*l.c.*) cites it from cacao and *Cola* sp. in the Cameroons. In more recent times, Mayné and Ghesquière (*l.c.*) record this species as being very common at Mayumbe on the twigs and pods of cacao, which it covers to such an extent that the parasitised fruits are not able to reach maturity. This latter conclusion can be confirmed from the writer's experience, though no numerical data are available to show the degree of damage caused. It is almost invariably attended in the field by the red tree ant, *Oecophylla smaragdina*, F., though the writer has also taken three species of *Crematogaster* and one *Pheidole* attending it. Other West African records include: cacao at Kpandu, B. M. Togoland (H. E. Box), pigeon pea at Aburi and plantain (*Musa paradisiaca*) at Tafo (G. S. Cotterell), *Calophyllum inophyllum*, *Glyphaea lateriflora*, *Musanga smithii* and *Loranthus bangwensis*, at Tafo (E. O. Boafo), and *Cola nitida* in Sierra Leone (Hargreaves, *l.c.*). Lamborn (1914) cites the larvae of *Eublemma ochrochnoa*, Hmps. and *Tortrix callopista*, Durrant, as predacious on this scale at Moor Plantation, Ibadan, Nigeria.

(27). *Geococcus coffeae*, Green, 1933.

This species is very common on the roots of cacao, *Canna indica*, *Coffea arabica* and *Desplatzia dewevrei* at Tafo. The species was first described from the roots of *Coffea liberica* at Surinam, Dutch Guiana, and was for a time considered a neotropical species. Green, in the type description, states that the specimens may be immature adults and gives the size as 1·5 to 2·0 by 0·5 mm. The writer has taken a number of undoubtedly adult females, in association with first-instar larvae, and these have measured (when mounted for microscopical examination), between 2·0 and 2·5 mm. long, and 0·8 mm. wide. They were elongate to oval in shape, covered with a thin coat of powdery meal, thinner along the mid-venter, beak, coxae and intersegmental membranes. The insects form small cells, lined with white wax, along the rootlets on which they are feeding. The writer has looked for this species on cacao roots in two other localities in the Eastern Province (Akwadum and Dawa) but has failed to find them. It should be mentioned that the above is the first African record of this very characteristic species.

(28). *Coccidella spelaea*, **sp.n.** (Figs. 1–6).

Adult Female (fig. 6) oval in shape, distinctly segmented, especially with regard to the abdominal segments. Covered with a thin layer of finely particulate white wax, thinner along the intersegmental membranes and around the beak and coxae. With one pair of short, stout, wax filaments on the last abdominal segment. Body colour, when wax removed, a very pale brownish yellow. *Antennae* (fig. 1) 6-jointed, basal joints of the two antennae separated by a gap equal to about the width of one. First and second joints as broad as long; third joint longer than broad; fourth and fifth joints as broad as long; sixth joint half as broad as long. The first five joints all have subapical rings of 4–5 setae. The fifth joint has, as well, one stout, gently curved, subapical spine, and the sixth joint has median, subapical, and apical rings of setae, and a tuft of 2–3 setae at the tip; in addition it has one median, and two subapical bluntly pointed, curved spines. Relative lengths of the joints, in microns, are as follows: (1) 30; (2) 19; (3) 25; (4) 19; (5) 19; (6) 46. *Hind legs* (fig. 2) with coxa robust, slightly broader than long, with a few setae along the integumental edge; trochanter large and robust, with one pair of long setae on the distal edge, and 2–3 setae on the proximal edge; femur one third as broad as long, with four rows of setae, one on each edge, and one medially on either side; tibia nearly three times as long as broad, with four rows of setae, these being more than half as long as the joint; tarsus tapering, at the base less than half as broad as long, at the distal extremity less than a quarter as broad as long, with four rows of setae, nearly three-quarters the length of the joint; tarsal digitules absent; claw very long, thin, gently curved, and sharply pointed; ungual digitules consisting apparently of a very fine hair, equal to less than half the claw in length. Measurements, in microns, of the joints of the legs are given below.



FIGS. 1–6.—*Coccidella spelaea*, sp. n.: 1, antenna; 2, leg; 3, anal cerarius; 4, anal ring; 5, ventral circulus; 6, adult female.

	Coxa		Femur		Tibia		Tarsus		Claw
	Length	Breadth	Length	Breadth	Length	Breadth	Length	Breadth	Length
Leg 1 ...	53	53	76	38	57	19	55	19	30
Leg 2 ...	49	53	80	35	53	20	53	19	30
Leg 3 ...	38	46	95	38	72	19	57	19	30

Cerarii (fig. 3) anal pair only present. These consisting of three or four long. thin setae, approximately 120 microns long, a few small circular pores, and 6–7 short setae, on raised and definitely sclerotised papillae on either side of the anus. Beak, much reduced in size; dimerous. Anal ring (fig. 4) strongly sclerotised, with 6 long, thin setae, approximately 100 microns in length, and with two rings of irregular, elongate, oval pores, those in the inner ring being slightly larger than those in the outer ring. *Dorsal osteoles* : two pairs present, with thickened lips, apparently without setae, but with a few (one or two) circular pores. Eyes : absent. Cephalic plate : absent. *Derm* : distinctly segmented, with a few short setae and circular pores on each segment, the dorsum being more heavily setose than the venter, and the anterior abdominal and thoracic segments being, ventrally, almost bare of setae. On the venter there are three circuli, one on the last thoracic segment, and one on each of the first two abdominal segments. The circulus on the first abdominal segment is the largest. The circuli are saucer-shaped (fig. 5) with the edges lightly sclerotised, and the lumen faintly striated with a mosaic pattern. There are also a number of monolocular tubular ducts on the venter. These vary in numbers, from, for example, 42 distributed variously over the segment, to 7 clustered on each edge, with 9 more or less in a line, connecting the two clumps, across the mid venter, or 6–7 clustered round the penultimate circulus, with 4 at either edge of the segment. Tritubular cerores present around the anterior margin, and on the venter in small numbers only. They are very small in size, and hard to find. An examination of the available material has shown 5–6 to be present around the anterior margin, from just behind the bases of the antennae, with 1 between the antennae ; about 10 present on the venter posterior to the beak, and between the first two pairs of coxae, with 1 only present on the venter anterior to the beak.

Length 2·0 mm. ; breadth 1·32 mm.

TYPE LOCALITY : Tafo, Eastern Province, Gold Coast.

TYPE HOST : *Theobroma cacao*, L. on roots. Coll. : *E. O. Boafo*, 13/viii/45.

MATERIAL EXAMINED : Twelve adult, and young adult, females. Type slide deposited in the British Museum (Natural History) Coccid collection.

This species would seem to come midway between the genus *Rhizoecus*, Künckel and *Coccidella*, Hambleton, as defined by Hambleton (1946; 1946a), in his recent revision of the hypogoeic mealybugs. It differs from all the species of *Coccidella* by being totally blind and yet has antennae that are definitely 6-jointed, and the tritubular cerores are by no means conspicuous or easy to find. It differs from all of the species at present assigned to *Rhizoecus* by its 6-jointed antennae and the scarce nature of the tritubular cerores. An examination of the slides of the species in the British Museum collection assigned by Hambleton to *Coccidella* has, however, shown that the present species is probably more closely congeneric with them than with the true species of *Rhizoecus* and so, for the present at any rate, it has been assigned to *Coccidella*.

(29). *Pseudococcus bukobensis*, Laing, 1929.

This species has been found to be quite common on cacao in the Gold Coast. On the first occasion on which it was taken, it was infesting, at major pest proportions, the branches and shoots of three young cacao seedlings, all growing within ten feet of the bole of a full-grown *Trema guineensis* (Ulmaceae). The bole of the *Trema* was covered with white meal but no mealybugs could be found on it. All of the specimens on the surrounding cacao were nymphs or young adults. A month later, 38 cacao seedlings, all within 40 yards of the *Trema* were infested with mealybugs. During this time, the mealybugs on the first three seedlings had become mature, and were seen migrating back over the litter and up the trunk of the *Trema*. After their return, the adults started producing ovisacs of loosely woven strands of white wax containing numerous minute yellow eggs. Two days later, torrential rains destroyed the infestation and no information could be gained on the length of oviposition or hatching time. The adult females were covered entirely with a thick layer of white wax, thinner along the segmental membranes, and ventrally around the beak and coxae. Without a median dorsal stripe. With 17 or 18 pairs of wax filaments, short, stout, with the anal pair a little longer than the rest. Body colour, when wax removed, a flesh-pink. This species has also been taken from cacao at Aburi, Akwadum, Anyboni, in the Afram Plains, at Kpeve in British Togoland and at Asokore and Olanla in Nigeria, and on two occasions has been found breeding on cacao. It has also been collected from Annonaceae sp., *Sterculia tragacantha*, *Ficus asperifolia* and *Ananas comosus*, and 6 species of ants (*Crematogaster* (2), *Pheidole* (3) and *Camponotus* (1) have been taken " attending " the colonies. The favourite habitat would seem to be in cracks in the bark of the host plant. Larvae of an undetermined Tortricid moth, and a Coccinellid beetle, have been taken at Tafo actively predacious on all stages of this mealybug and one Encyrtid parasite has been bred from adult females.

(30). *Pseudococcus concavocerarii*, James, 1934.

In the type description, James states that the " . . . anal, penultimate, and antipenultimate pairs of tassels may be longer than twice the length of the body; and the lateral tassels may exceed in length the width of the body." In all of the Gold Coast material seen by the writer, the lateral wax tassels were about half the body width, the posterior and anal pairs being 3–6 times as long as the laterals. Live specimens, with the wax brushed away, were a dull brownish grey in colour.

This species occurs sporadically on cacao throughout the Eastern Province and has been taken ten times at Tafo in the last year. It rarely occurs on cacao in large numbers, 3–4 young adults on a pod, or perhaps a dozen on a young shoot, being usual. Much heavier infestations have been noted on *Jasminum grandiflorum*, *Hippeastrum* sp., *Codiaeum* sp. and *Aristolochia* sp. Mature specimens, mounted on a slide, measured 2·5 to 3·0 by 1·8 to 2·0 mm. All stages are very active if disturbed. This species was described from *Coffea arabica* at Kitale, Kenya Colony.

(31). *Pseudococcus citri* (Risso, 1813).

?

This species, described from *Citrus* sp. from France, is so widespread and well known that it needs little introduction to modern entomologists. The following points may, however, be of some use in separating it from the other West African cacao mealybugs. Elongate oval in shape, with 17 pairs of short, stout, wax filaments forming a lateral fringe. The anal, and two preceding, pairs, slightly longer than those laterally placed. Body covered with a layer of finely particulate white wax, with a conspicuous, but faint, stripe on the dorsum, extending from the first thoracic to the mid-abdominal segments. This stripe is caused by a much decreased wax deposit in the middle of the segments concerned. Body colour, when wax removed, is a characteristic pale yellow.

P. citri has been incriminated by Box (1945) as a vector of the strains C and D of the Swollen Shoot virus complex, though these results have yet to be confirmed. Posnette has recently informed the writer (*in litt.*) that he has now obtained transmission of the strain F with this mealybug. *P. citri* is very widely distributed throughout the West African cacao belt and especially so in Nigeria. The writer has examined material from 22 localities in the Eastern Province, 5 in Ashanti, Gold Coast, 3 in British Togoland and 16 in Nigeria, as well as many collections from various food plants at Tafo. As a whole, these collections exhibit surprisingly little variation, the main factors of variance being the numbers of spines in the cerarii (a few specimens have 1 or 3 spines in a cerarius, instead of the usual 2) and the size of mature females (some specimens measuring 2·5 mm. when mounted on a slide, others perhaps only 1·8 mm.). One specimen was taken which showed an extraordinary abnormality—a " forked " antenna, in which the 7th and 8th joints were duplicated, and jointed to the 6th joint in the usual manner, forming a Y-shaped organ. The identity of this species has been settled by Professor Ferris, in a letter to H. E. Box, written in 1944, and further material was subsequently similarly identified by Dr. Hall, of the Imperial Institute of Entomology, and there can be little doubt that it is the true *P. citri* that is present. Twenty species of ants have been taken in the field "attending" *P. citri* colonies on cacao and other plants. These include *Crematogaster* (*Sphaerocrema*) *luctans*, Forel, *Crematogaster* (*Atopogyne*) ? *kneri*, Mayr, eight other undetermined species of *Crematogaster*, nine species of *Pheidole* and one of *Camponotus*. A list of the 52 species of food plants, apart from cacao, from which *P. citri* has been collected in the Gold Coast by Box, Cotterell, the writer, and his associates and assistants is given below.

A. DICOTYLEDONS.

Annonaceae. *Annona squamosa*, *Uvariodendron* sp.
Piperaceae. *Piper umbellatum*, *P. guineense*.
Samydaceae. *Homalium dolichophyllum*.
Combretaceae. *Terminalia superba*.
Tiliaceae. *Cistanthera papaverifera*, *Desplatzia lutea*, *Glyphaea lateriflora*, *Triumfetta* sp.
Sterculiaceae. *Sterculia rhinopetala*, *S. tragacantha*, *Cola acuminata*, *C. togoensis*, *Erythropsis barteri*.
Bombacaceae. *Ceiba pentandra*.
Euphorbiaceae. *Mallotus oppositifolius*, *Macaranga* sp.
Mimosaceae. *Albizzia* sp.
Papilionaceae. *Baphia nitida*, *Amphimas pterocarpoides*, *Gliricidia maculata*.
Moraceae. *Ficus asperifolia*, *Ficus* sp., *Treculia africana*.
Urticaceae. *Fleurya aestuans*.
Ampelidaceae. *Cissus* sp.
Anacardiaceae. *Mangifera indica*.
Connaraceae. *Cnestis ferruginea*.
Sapindaceae. *Deinbollia pinnata*, *Blighia sapida*.
Sapotaceae. *Malacantha* sp.
Rubiaceae. *Coffea arabica*, *C. liberica*, *Canthium glabriflorum*, *Randia genipaeflora*, *R. maculata*.
Compositae. *Vernonia conferta*.
Solanaceae. *Datura metel*.
Bignoniaceae. *Kigelia africana*.
Acanthaceae. *Brillantaisia nitens*, *Brillantaisia* sp.
Verbenaceae. *Clerodendron formicarum*.

B. MONOCOTYLEDONS.

Commelinaceae. *Palisota thyrsiflora*, *Commelina capitata*.
Bromeliaceae. *Ananas comosus*.
Musaceae. *Musa paradisiaca*.
Zinziberaceae. *Costus afer*.
Marantaceae. *Clinogyne leucantha*.
Araceae. *Culcasia angolensis*, *Xanthosoma sagittifolium*.
Amaryllidaceae. *Haemanthus multiflorus*.
Dioscoraceae. *Dioscorea* sp.

In addition to these plants, Hargreaves (*l.c.*) cites *Anisophyllea laurina* as a food plant in Sierra Leone.

The commonest parasite of *P. citri* in the Gold Coast is a small red Cecidomyiid that, in the larval stages, sucks the body fluids of the host. On maturity, the larvae pupate underneath the dead bodies of the host insects. On several occasions the mealybugs have been seen to lay some eggs before succumbing to the depredations of the parasites.

(32). *Pseudococcus hargreavesi*, Laing, 1925.

This species, described from *Bauhinia* sp. from Kampala, Uganda, has been collected twice on cacao by Box, once at Nkawkaw, Gold Coast, and once at Ikire, Nigeria. It has never been encountered by the writer and must be a rare species.

(33). *Pseudococcus longispinus*, Targioni-Tozzetti, 1867.

This species has been taken by Mayné and Ghesquière (*l.c.*) from cacao, cotton, *Hevea* rubber, *Cassava*, *Poinsettia* sp., *Jatropha multifida*, *Bidens pilosa* and *Poinciana regia*, in the Congo. Cotterell has taken it from *Ceiba pentandra* at Kpeve, Togoland. Other African records include Smee (1940) from cotton in Nyasaland,

and Lamborn (1914) from cacao at Ibadan, Nigeria. (In the latter case, the identity is open to doubt.) Mayné and Ghesquière cite two Cucujid beetles, *Laemophloeus janeti*, Grouv., and *L. nebulosus*, Grouv., as predators in the larval stages.

(34). *Pseudococcus njalensis*, Laing, 1929.

This species is undoubtedly the commonest cacao mealybug in the Gold Coast. It is almost certainly the chief natural vector of the virulent Strain A Swollen Shoot virus, and as such is of great economic importance. It was originally described by Laing from Sierra Leone on coffee; later (1944) the same author described *P. exitiabilis* from the Gold Coast on cacao. Hall (1945), however, after examining an extended series of material, came to the conclusion that a single variable species existed, and that the specific name *exitiabilis* should sink as a synonym of the earlier *njalensis*. The morphology of the adult female has been adequately covered by these two authors. A field description of the female is as follows :—

With 18 pairs of lateral wax filaments of medium length, the anal pair longest, the 17th and 16th pairs a little shorter, but longer than those placed anteriorly and laterally. Body covered with mealy secretion, thinner along the intersegmental membranes, and around the base of the beak, coxae, and mid-venter, and, in many cases, anteriorly dorsal to the lateral filaments; this secretionary matter is absent in a streak along the mid-dorsal line, the free areas being widest and broadest on the 2nd and 3rd thoracic segments, and tapering off gradually on the anterior abdominal segments. Body colour, when secretionary covering has been removed, light chocolate brown dorsally, merging into a flesh pink ventrally. Some specimens are a uniform flesh pink in colour, whilst others have been either a light yellow or dark reddish brown. This species can be distinguished from *P. citri* in the field most readily by its more nearly circular shape, the more prominent, but shorter, mid-dorsal streak, and the body colour under the secretionary covering. Length of freshly killed, but unmounted, material, approximately 2·0 mm., breadth, 1·8 mm.

Nearly 50 species of ants have so far been taken attending *P. njalensis* colonies in the field. It is the writer's opinion that this trophobiotic association is of greater benefit to the ants than to the mealybugs. It is true that the ants build carton tents over the mealybug colonies, thus protecting them from inclement weather conditions, and they have been seen on two occasions by the writer actually transporting young nymphs from colonies on felled trees, and establishing them on nearby living trees (on neither occasion were the mealybugs carried more than 6 feet), and they also keep the bugs free of the grey mould (a mixture of species, with an *Aspergillus* predominating), which rapidly infects them if the sugary secretions are not removed. On the other hand, several of the species of ants concerned have been seen eating the larval mealybugs. In a small scale laboratory experiment, seven cacao seedlings were infested with mealybugs and ants, and seven with mealybugs only, and, the mealybug populations being identical at the start of the experiment, the colonies were allowed to develop naturally for 6 months. At the end of this time, the ant-free plants all had far higher mealybug populations than the ant-attended plants, though the "no-ant" colonies were covered with mould, and were dispersed all over the plants in question. The " ant " plants were free of mould, and the colonies remained compact. On every occasion that *P. njalensis* has been taken in the field, it has been attended by ants. In spite of this, it is often parasitised as much as 12 per cent. by the small red Cecidomyiid mentioned in connection with *P. citri*, and two other Hymenopterous parasites, *Neodiscodes martinii*, Comp., and ? *Chiloneurus* sp. (determined by G. Nixon) have been bred from it. Two as yet undetermined Lepidoptera are, in the larval stages, predacious on *P. njalensis*, as well as the larvae of at least one species of Coccinellid, and one Staphylinid, beetle. The ants seemingly pay no attention to the predators. Only nine of the ant species have so far been identified: *Crematogaster* (*Sphaerocrema*) *boxi*, Donis., *C.* (*S.*) *striatula*, Emery, *C.* (*S.*) *luctans*, Forel, *C.* (*Atopogyne*) ? *kneri*, Mayr, *C.* (*A.*) *africanum*, Mayr, *C.* (*A.*)

cuveriae, Donis., *C.* (*Nematocera*) *stadelmanni*, Mayr, *Meranoplus nanus*, Andre, and *Monomorium pharaonis*, L. The last named species would seem to be only incidentally coccidophilic, and has only been taken attending *P. njalensis* colonies on cacao pods brought into the laboratory for experimental purposes. *P. njalensis* has been taken from nearly 100 different species of food plants apart from cacao. Hall (1945) has already cited 21 food plants, and the others are given below.

A. DICOTYLEDONS.

Annonaceae. *Enantia chlorantha, Uvariodendron* sp., Annonaceae sp.
Lauraceae. *Persea gratissima.*
Polygalaceae. *Carpolobia lutea.*
Cucurbitaceae. *Telfairea occidentalis.*
Lecythidaceae. *Napoleona parviflora, Combretodendron africanum.*
Tiliaceae. *Desplatzia lutea, D. dewevrei, Ledermannia chrysochlamys.*
Sterculiaceae. *Cola acuminata, C. cordifolia, C. chlamydantha, C. ? lateritia, Sterculia tragacantha, S. elegantifolia.*
Euphorbiaceae. *Antidesma laciniatum, Microdesmis puberula, Ricinodendron africanum, Macaranga barteri, M. heudolotii, Codiaeum* sp.
Caesalpiniaceae. *Hymenostegia afzelii, Chidlowia sanguinea, Cassia siamea.*
Mimosaceae. *Acacia pennata, Albizzia ferruginea, Parkia filicoidea.*
Papilionaceae. *Erythrina* sp., *E. addisoniae, Baphia nitida, Lonchocarpus* sp.
Ulmaceae. *Trema guineensis.*
Moraceae. *Musanga smithii.*
Rutaceae. *Fagara xanthoxyloides.*
Sapindaceae. *Paullinia pinnata, Phialodiscus unjugatus, Blighia sapida.*
Anacardiaceae. *Anacardium occidentale.*
Araliaceae. *Cussonia* sp.
Sapotaceae. *Synsepalum dulcificum.*
Apocynaceae. *Carissa edulis, Motandra guineensis, Conopharyngia* sp.
Asclepiadaceae. *Pergularia extensa.*
Rubiaceae. *Coffea liberica, Craterispermum cerinanthum, Randia malleifera, Mitragyna stipulosa, Mitragyna* sp., *Psychotria* sp.
Compositae. *Vernonia conferta, Aspilia latifolia.*
Solanaceae. *Solanum torvum.*
Bignoniaceae. *Newbouldia laevis.*
Acanthaceae. *Brillantaisia nitens.*
Verbenaceae. *Vitex ? grandifolia, Vitex* sp., Verbenaceae sp.
Labiatae. *Platystoma africana.*

This list contains some records made by H. E. Box as well as those made by the writer. In addition, Hargreaves (*l.c.*) cites *Cola nitida* as a host plant in Sierra Leone.

(35). *Ferrisia virgata* (Cockerell, 1893).

This species transmits the Strain A Swollen Shoot virus and, were it as common as *P. njalensis* in the main cacao areas, would probably be an equally important field vector. In the Gold Coast, the adult female is thickly covered with white meal dorsally, not quite so thickly ventrally, with three pairs of wax free spots on either side of the mid-dorsal line, as follows: first pair, " pin's head size," on 2nd thoracic segment; second pair, same size as first, on 3rd thoracic segment; third pair, triangular in shape, pointed posteriorly, 5–6 times greater than the thoracic spots in size, on the anterior abdominal segments. Body covered with thin, glassy, wax filaments dorsally and laterally. No lateral fringe of wax tassels. One pair of wax filaments anally, these being ½–¾ as long as the insect. Body colour, when wax removed, dark blue-black dorsally, greyish ventrally. Size, just before depositing ovisac, 4·0 by 2·0 mm. *F. virgata* seems to be rare in the majority of " bush " cacao farms, but common wherever introduced ornamental and garden plants are to be found, and particularly so in the dry areas, such as the Accra plains. On a number of occasions the writer has seen it colonising bushes along main roads, and it would seem that its spread has coincided more or less with that of certain introduced plants, in particular *Acalypha* spp. and *Leucaena glauca*. The following list gives the species of plants, apart from cacao, from which *F. virgata* has been collected in the Gold Coast. It includes some records made by Cotterell and Box.

A. DICOTYLEDONS.

Annonaceae. *Annona squamosa.*
Piperaceae. *Piper guineense.*
Violaceae. *Viola* sp.
Portulacaceae. *Portulaca* sp.
Amarantaceae. *Cyathula prostrata.*
Geraniaceae. *Pelargonium* sp.
Combretaceae. *Terminalia superba.*
Tiliaceae. *Cistanthera papaverifera.*
Sterculiaceae. *Sterculia tragacantha, Cola* sp.
Malvaceae. *Hibiscus rosa-sinensis, H. mutabilis, Gossypium* sp.
Euphorbiaceae. *Macaranga heudolotii, Ricinodendron africanum, Acalypha* spp., *Manihot* spp., *Jatropha* sp.
Caesalpiniaceae. *Caesalpinia crista.*
Mimosaceae. *Acacia pennata, Leucaena glauca.*
Papilionaceae. *Sophora tomentosa, Amphimas pterocarpoides, Erythrina* sp., *Desmodium ? abyssinica.*
Casuarinaceae. *Casuarina equisetifolia.*
Urticaceae. *Urera* sp., *Fleurya* sp.
Rubiaceae. *Mitragyna stipulosa, Canthium glabriflorum.*
Solanaceae. *Datura metel.*
Verbenaceae. *Duranta repens.*

B. MONOCOTYLEDONS.

Commelinaceae. *Commelina nudiflora.*
Musaceae. *Musa paradisiaca.*
Marantaceae. *Clinogyne leucantha.*
Cannaceae. *Canna indica.*
Araceae. *Xanthosoma sagittifolium.*

In addition to these plants, artificial infestations were satisfactorily established on *Sterculia rhinopetala, Cola acuminata, C. cordifolia* and *Triplochiton scleroxylon* (Sterculiaceae) and on *Bombax buonopozense* and *Ceiba pentandra* (Bombacaceae). Other West African records for this species include *Arachis* sp., Avocado pear, *Bauhinia* sp., *Crescentia* sp., *Calopogonium mucunoides, Cassia sieberiana, Clerodendron paniculatum, Coffea* sp., *Gliricidia maculata, Psidium guajava, Tecoma stans, Waltheria lanceolata* and cultivated tomato (*Solanum* sp.), all cited by Hargreaves (*l.c.*) from Sierra Leone. Newstead (1917a) records this species from cacao in Southern Nigeria. One Hymenopterous parasite, *Neodiscodes martini* (the same species that has been reared from *P. njalensis*), has been bred commonly from *F. virgata*. Two species of ants, both *Pheidole*, have been taken attending colonies of *F. virgata* in the field. *F. virgata* is not usually attended by ants, presumably because of the long glassy wax filaments, which deter most species, and it is possible that the ants were chance visitors at the time of collection.

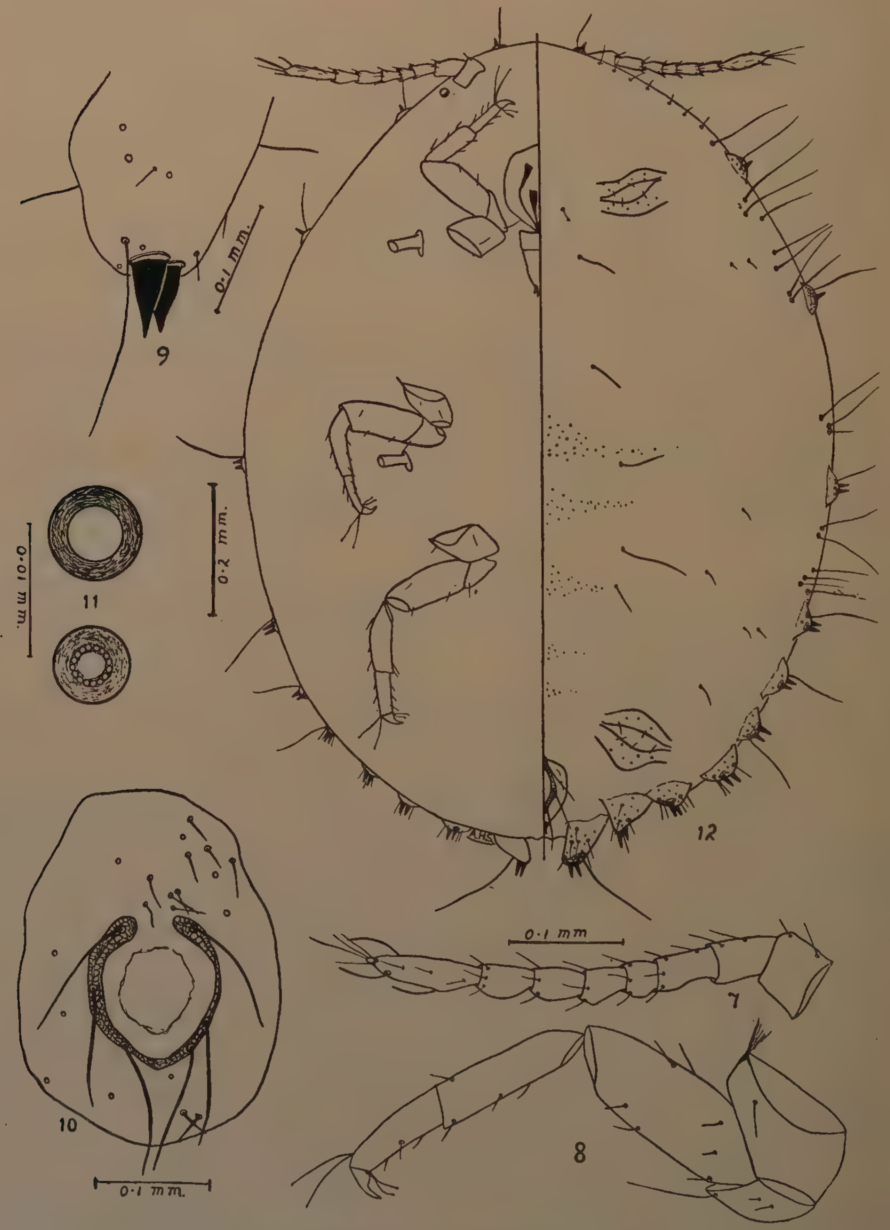
Not in

(36). *Ferrisia virgata* var. *madagascariensis* (Newstead, 1908b).

This variety was recorded from cacao in Southern Nigeria by Peacock (1913). It has not been seen by the writer in the Gold Coast.

(37). *Tylococcus westwoodi*, **sp. n.** (Figs. 7–12).

Adult Female (fig. 12) oval in shape, narrowed anteriorly. Body colour apparently dark red, but material preserved two days in alcohol before examination, so no field description is possible. Specimens with a few strands of a dark red wax still adhering to the dorsum. *Antennae* (fig. 7) 8-jointed, the first three joints equal in length, the fourth shortest, the fifth and sixth equal in length, and a little longer than



Figs. 7–12.—*Tylococcus westwoodi*, sp. n.: 7, antenna; 8, leg; 9, anal cerarius; 10, anal ring; 11, derm pores; 12, adult female.

the fourth, the seventh equal to, or a little longer than, the third, the eighth longest, and equal to the sixth and seventh together. The first seven joints each with a subapical ring of 3–4 setae, the setae being about half as long as the basal joint. The seventh joint also has one thick, stout, curved, subapical spine. The eighth joint has a medium ring of 2–3 short setae, a tuft of short setae at the apex, and three stout spines, one medially and two subapically placed. Relative lengths of the joints, in microns, are: (1) 45; (2) 48; (3) 47; (4) 28; (5) 35; (6) 36; (7) 42; (8) 86. *Hind leg* (fig. 8) stout, and robust. Coxa, broader than long, with a few (1–2) setae along the edge. Femur, shorter than tibia and tarsus together, and twice as broad as tibia, with not more than 6–8 setae, mostly along the inner edge. Tibia and tarsus also with most of their setae ranged along the inner edge. Tarsus with a pair of digitules, ¾ length of tarsus. Claw gently curved, one-third length of tarsus, with a pair of knobbed digitules slightly longer than claw. Relative measurements of the joints, in microns, are given below: —

	Coxa	Femur	Tibia	Tarsus	Claw
Leg 1 ...	48	150	105	82	27
Leg 2 ...	51	157	113	84	27
Leg 3 ...	48	164	121	87	27

The third pair of legs are slightly longer than the other two pairs. *Cerarii* (fig. 9) usually eleven, sometimes ten or twelve pairs, all on raised, sclerotised, tubercles. The anal tubercles largest (90 by 79 microns), the remainder progressively smaller in size. Each tubercle with two (or, anteriorly, only one) strong spines at apex, a cluster of circular pores, and a few short setae. Anal pair with one long seta, and lateral and anterior pairs with one or two long setae. Beak, obscurely dimerous, 2½ times as long as broad, with a cluster of fine hairs at the tip, and 2–3 medially placed on terminal segment. Anal ring (fig. 10) with two rows of pores and six long, relatively stout, setae; area surrounding with a few pores and setae, mostly situated dorsal to orifice, and with one pair of short, sharp spines ventral to the orifice. *Dorsal osteoles* conspicuous, each with a few circular pores, and a few short setae on the thickly sclerotised lips. Eyes circular, flattened, saucer-like. Spiracles large and conspicuous. *Derm* obscurely segmented, with clusters of thick-lipped circular pores (fig. 11) on the mid-dorsum, and scattered over dermis. Mingled with these scattered pores are some slightly smaller, and apparently multilocular, pores. A few setae, either short or long, scattered on dorsum, the long ones (2–3 times as long as the short ones) more or less restricted to the lateral margins, and often in clusters near a cerarius. Pores and setae sparse on venter.

Length 1·5 mm., breadth 1·2 mm.

Type Locality: Atikplale, Eastern Province, Gold Coast.

Type Host: *Theobroma cacao*, small colony on a pod, attended by an undetermined ant of the genus *Crematogaster*. Coll.: *D. Westwood*, to whom this species is dedicated.

Material Examined: Fourteen adult and young adult females. Type slide deposited in the British Museum.

This species is assigned to *Tylococcus*, Newstead, because of the small, but definite, sclerotised tubercles that form the bases of the cerarii. The writer has recently had an opportunity of examining the genotype, *Tylococcus madagascariensis*, Newst. (1897), and there can be no doubt as to the congeneric nature of the new species. It differs from *T. madagascariensis* in only having eleven pairs of cerarii, and in the size of the tubercles (smaller), and the cerarian spines (larger), than the genotype. It should be pointed out that many of the species that have been, in the past, assigned to *Tylococcus* have subsequently been placed in other genera (*T. formicarius*, Green and *T. simplicior*, Green, for example, have been put into *Farinococcus*, Morrison (Laing, 1925)). The writer has recently examined a slide of *T. giffardi*, Ehrhorn, in the British Museum collection, and can state that this species is certainly not a true *Tylococcus*, and must definitely be assigned to a new genus.

T. westwoodi has so far been taken only once, and, unless it turns out to be a vector of the Swollen Shoot virus, can be considered of no economic importance.

(38). *Paraputo ritchiei*, Laing, 1929.

P. multispinosa, James (syn. nov.).

This species has been collected nine times from cacao and other plants in the last year. Before proceeding with the biological notes, it would be advisable to clear up one or two points regarding the taxonomy of the species. The only other member of the genus (*multispinosa*, James) was described in 1935 from an undetermined ornamental plant at Kitito, Ukamba Province, Kenya. In his type description, James states: "The main feature distinguishing this species from *P. ritchiei*, Lg., the only other member of the genus, is the markedly greater number of conical spines present on the posterior six pairs of cerarii. . . ." He then gives the following table :—

	P. multispinosa	*P. ritchiei*
Last pair of cerarii ...	8–11	7
2nd last „ „ ...	20–24	15–17
3rd „ „ „ ...	23–26	15–17
4th „ „ „ ...	16–27	15–17
5th „ „ „ ...	18–22	11–13
6th „ „ „ ...	13–15	11–13

He continues: "Other significant differences are the absence of translucent pores on the hind femora, and their restriction to the basal region of the hind coxae in *P. ritchiei*; also a bunch of from 5 to 7 rather long and stout setae which occurs on the postero-dorsal aspect of each paranal lobe in *P. multispinosa* is absent in *P. ritchiei*." An examination of 30 specimens from eight of the nine collections received by the writer has shown that most of James' differences collapse when an extended series are compared together, and with the types of both species. For example, in collection H.5527, out of the 8 specimens examined, 2 had 10 pairs of cerarii, 1 had 11, 3 had 13, and 2 had 14 pairs. The number of spines in the six posterior pairs, starting with the anal pair, was: (1) 7–13, mean: 11; (2) 11–26, mean: 20; (3) 13–25, mean: 17; (4) 13–28, mean: 18; (5) 11–21, mean: 14; (6) 8–16, mean: 11. In collection H.5421, out of the 9 specimens examined, 3 had 9 pairs of cerarii, 2 had 10, 2 had 11, 1 had 12, and 1 had 13, pairs. Spine counts were: (1) 9–14, mean: 11; (2) 15–25, mean: 20; (3) 11–23, mean: 18; (4) 12–20, mean: 15; (5) 11–17, mean: 14; (6) 8–13 mean: 10. In collection H.5528, the 8 specimens gave the following counts: 1 specimen had only 8 pairs of cerarii, 6 had 9 pairs, and 1 specimen had 10 cerarii on one side, and 9 on the other. Counts were: (1) 11–15, mean: 12; (2) 15–21, mean: 19; (3) 14–23, mean: 18; (4) 15–20, mean: 18; (5) 11–17, mean: 13; (6) 7–15, mean: 11. It is clear that, so far as the numbers of spines in the cerarii are concerned, a great deal of overlapping ensues, not only amongst different colonies, but also amongst individual specimens in any one colony. Furthermore, the other characters detailed by James do not appear to be constant enough to be of specific importance. In collection H.5152, for example, one anal cerarius contains 7, and the other 8 spines, yet the paranal lobes bear 4 and 3 long, stout, setae respectively. In H.5057, carrying 9 and 11 anal cerarial spines (sufficient to assign it to the *multispinosa* species), there are no such setae on the paranal lobes. It would, therefore, seem as if a single variable species exists, and it is herewith proposed that *Paraputo multispinosa*, James, is sunk as a synonym of *P. ritchiei*, Laing. In the type description, Laing gives no field description of *P. ritchiei*. Gold Coast specimens appear as follows: Sub-circular in shape, mature specimens more nearly circular than young adults. Covered with white wax, of a lumpy nature, the lumps thicker and larger at the lateral edges of the segments. With 12 pairs of lateral wax filaments, all on the abdominal segments. Anal pair longest, next two pairs a little shorter, and then tapering off until the twelfth pair, which are approximately one-eighth as long as the anal pair. Body colour, when wax removed, flesh pink, darker dorsally than ventrally.

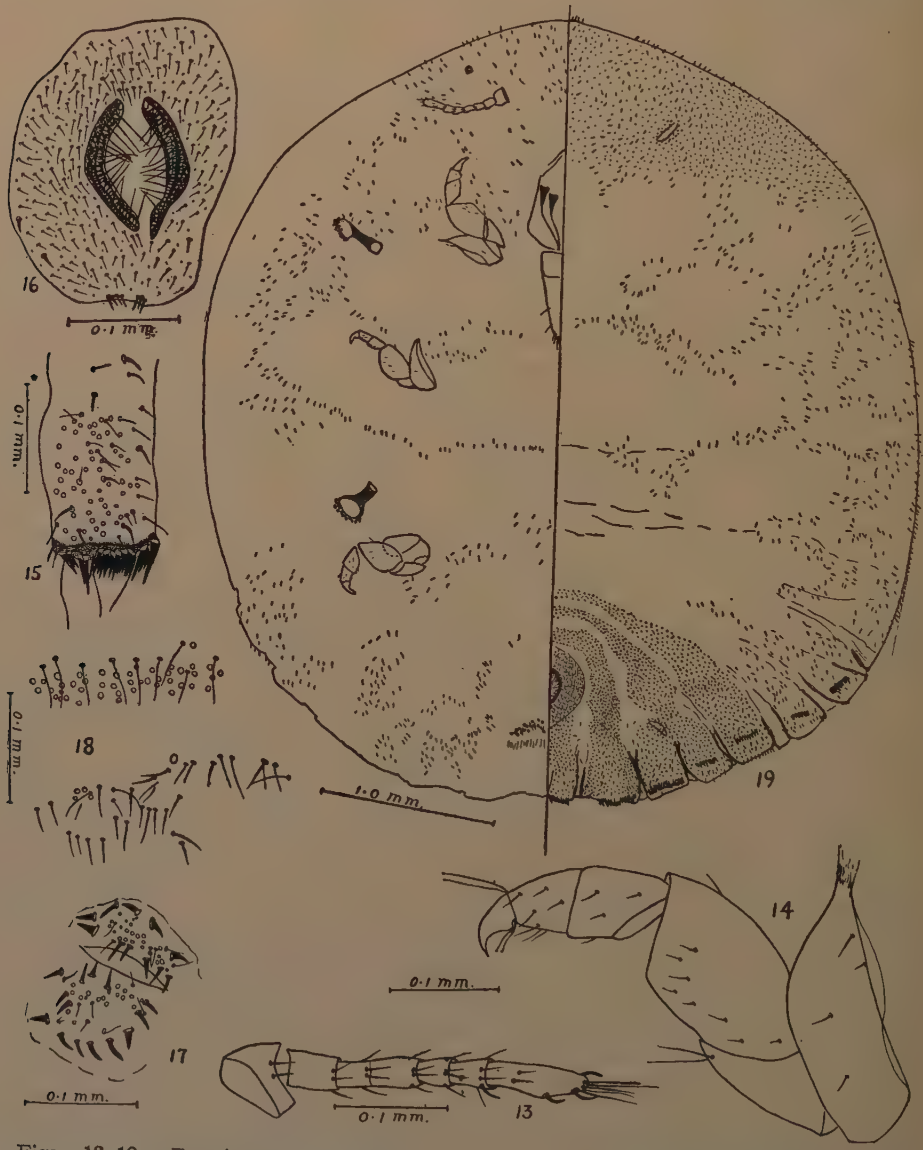
Paraputo ritchiei has been taken nine times in the past year. Once from cacao at Tafo and once at Kwabeng (Eastern Province), feeding in cracks in the bark, once from *Clinogyne cuspoidata* at Abompe and 6 times from Tafo, on *Cola cordifolia*, *Triplochiton scleroxylon* (twice), *Malacantha* sp., *Trichilia prieuriana* and *Chlorophora excelsa*. It invariably feeds in cracks and crevices in the bark of the host plant, often 15–20 feet above ground level.

(39). *Formicococcus tafoensis*, **sp. nov.** (Figs. 13–19).

Adult Female (fig. 19) circular to sub-circular in shape, usually narrower anteriorly than posteriorly. Covered completely with white wax, of a powdery consistency, easily removed with a soft brush. Twelve pairs of uniform square or rectangular wax plates laterally and posteriorly, and one pair of elongate, composite (*i.e.* seemingly consisting of 4–5 ordinary plates fused together) plates anteriorly. Wax thinner along intersegmental membranes, and around bases of beak and coxae. Body colour, when wax removed, pink ventrally, and a darker pink tinged with grey dorsally, changing rapidly to a uniform grey-black on death. *Antennae* (fig. 13) 7-jointed, although the division between the 3rd and 4th joints is often lacking or incomplete, giving the appearance of a 6-jointed antenna. Basal joint as broad as long, 2nd joint longer than broad, 3rd joint just over half as long as second, 4th joint longer than third, 5th joint same length as third, 6th joint same length as fourth, 7th joint twice as long as sixth. All joints with a sub apical ring of 3–4 setae. 6th joint with one curved, blunt, subapical spine in addition to setae, and 7th joint with submedian, and subapical setal rings, a tuft of 4 setae at the tip, and three blunt spines, one medially, and the others subapically placed. Relative measurements of the joints, in microns: (1) 38; (2) 51; (3) 29; (4) 42 (when joints 3 and 4 are fused, the single joint "3–4" measures 71); (5) 33; (6) 40; (7) 81 (fig. 13). *Hind leg* (fig. 14) much reduced in size, short, and stout. Coxa nearly three times as broad as long, with 1–2 setae along the integumental edge, and 1–2 on the median aspect. Trochanter with 2–3 setae on the distal corner. Femur more than half as broad as long, with 7–8 setae in two rows on the inner half, and only 1–2 on the outer edge. Tibia less than half length of femur, and nearly as broad as long, with a few setae on the inner edge. Tarsus shorter than tibia, nearly as broad as long proximally, tapering distally to just over half the length in width, with 6–7 setae along the inner edge and medially. Tarsal digitules very fine and thin, and about ¾ length of tarsus, with extremely fine knobbed ends. Claw ¾ length of tarsus, gently curved, stout, and robust. Ungual digitules relatively short and thin, apparently without, or with very finely, knobbed ends. Relative measurements, in microns, of the joints of the legs are given below.

	Coxa	Femur	Tibia	Tarsus
Leg 1	67	187	78	59
Leg 2	78	202	78	59
Leg 3	78	183	78	59

Cerarii (fig. 15) prominently present on the seven terminal abdominal segments. Spines long, thin, sharply pointed, present in varying numbers, in a broad marginal band on each segment. They are packed so closely together that they are very difficult to count accurately. However, the anal pair have approximately 20 spines each, and a few (3–4) long, thin setae, and clusters of circular pores intermingled with the spines and setae on the strongly sclerotised base. Subsequent pairs similar, but with more spines, the 7th (*i.e.* the foremost) pair with fewer spines. Means of four specimens gave the following counts (commencing with the anal pair and working forward)



Figs. 13–19.—*Formicococcus tafoensis*, sp. n.: 13, antenna; 14, leg; 15, anal cerarius; 16, anal ring; 17, dorsal osteole; 18, venter, genital region; 19, adult female. (Pores present on mid-segments, dorsally and ventrally, but omitted in order to show up spine clusters.)

(1) 21; (2) 49; (3) 46; (4) 43; (5) 35; (6) 26; (7) 13. In some specimens only 6 pairs of cerarii were present. Beak dimerous, with 4 pairs of short setae at tip, and 2 pairs of long setae (1½–2 times as long) medially and subapically placed. Anal ring (fig. 16) with 14–16 pairs of setae, with 2 rows of pores, the inner pore ring containing slightly larger pores than the outer ring; area surrounding the anus densely packed with pores and long, thin, setae, with, posteriorly, two groups of 3–4 sharp, relatively stout, curved spines. *Dorsal osteoles* (fig. 17) reduced in size, but with large lips crowded with spines and pores, and with a few short setae. The spines nearest the orifice are rather narrow at the base, and straight, those distally placed broader at the base, and usually markedly curved. Eyes small, circular, about the same diameter as the length of a body spine. Spiracles large and conspicuous, with clusters of circular pores around the openings. *Derm* clearly segmented, with deep intersegmental incisions posteriorly. Covered with circular pores dorsally and ventrally, the pores being interspersed with short, finely pointed, setae. The pores are distributed in relation to the segments, and are apparently all of one type dorsally, whereas ventrally an occasional larger pore is found, with a concentration of these larger pores on the penultimate abdominal segment. Derm covered irregularly with short, sharp, robust, gently curved spines, these being absent on the mid-dorsum except for two pronounced lines, and less common ventrally than dorsally and dorso-laterally. A band of long setae present on the venter posteriorly in association with the large pores (fig. 18).

Length 4·8 mm., breadth 4·6 mm.

TYPE LOCALITY: Tafo, Eastern Province, Gold Coast.

TYPE HOST: *Theobroma cacao.* Coll.: *E. O. Boafo.*

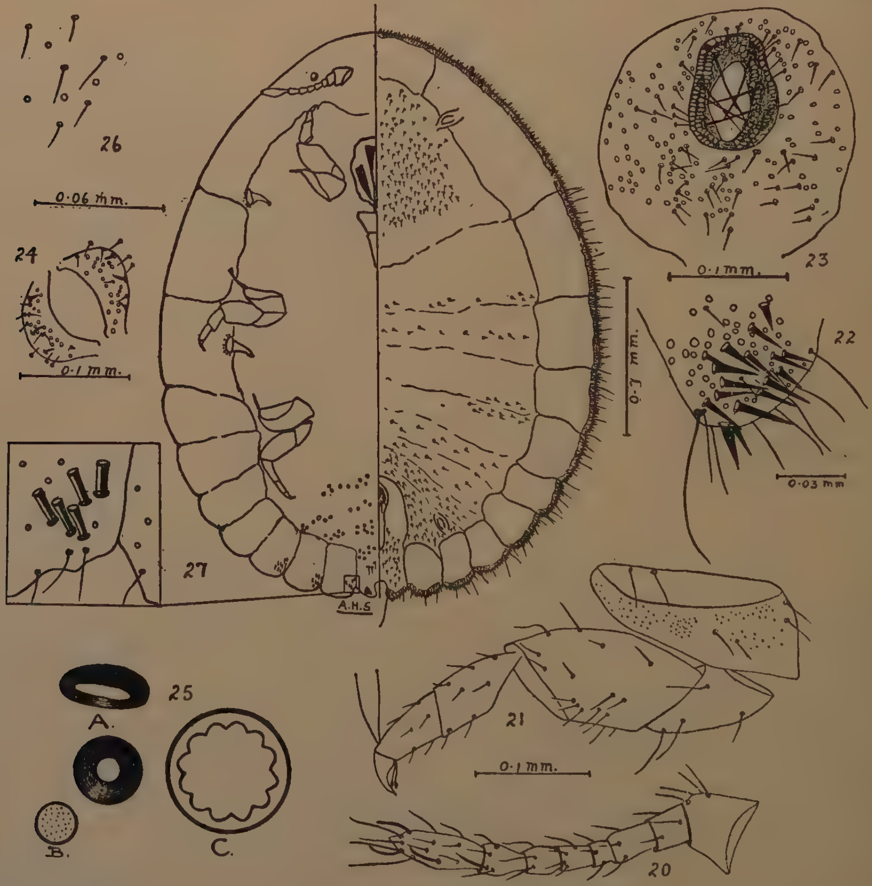
MATERIAL EXAMINED: Fifteen adult females. Type deposited in the British Museum collection.

This species has been assigned to the genus *Formicococcus*, Takahashi (1928), because of its undoubtedly close relationship with *F. humboldtiae*, Green, and *F. greeni*, Vays. It differs from *humboldtiae* in possessing deeply incised posterior abdominal segments, in the distribution of the dermal spines (which are very sparse in *humboldtiae*), and in the length of the third antennal joint, which is very much longer in *humboldtiae*, and (as in the new species) apparently fused to the fourth joint. It differs from *greeni* in the very reduced nature of the legs and antennae (which, in *greeni*, resemble more the normal Pseudococcine type), and in the number of anal ring setae (about 20 in *greeni*, and only 14–16 in the new species). It differs from *F. cinnamomi*, Takah., the genotype, in the number of setae in the anal ring, and in the fact that there is no continuation of the cerarii beyond the abdominal segments.

Formicococcus tafoensis has been collected from cacao twice, from *Ceiba pentandra* twice (once at Tafo, and once at Adownkwanta), once from *Terminalia superba*, once from *Cola cordifolia*, twice from *Alstonia congensis*, once from *Guarea cedrata*, twice from *Triplochiton scleroxylon*, all at Tafo, and once from *Casuarina equisetifolia*, at Kumasi, Ashanti. On every occasion it was taken feeding in cracks, or under flakes, of bark of the hosts, usually on buttress roots, or within 8 feet of the ground.

(40). *Farinococcus loranthi*, **sp. n.** (Figs. 20–27).

Adult Female (fig. 27) circular to sub-circular in shape, covered with a relatively thick coat of smooth white wax, except in the intersegmental areas, around the bases of the beak and coxae, and in two spots on the thoracic segments (one either side of the mid-dorsal line), and less thickly on the anterior abdominal segments in the mid-dorsal line. Postero-laterally there are 7 pairs of wax plates, the 4 pairs nearest the anus being rectangular in shape, and usually longer than wide, the next three pairs being wider than long. Anteriorly, this wax fringe is continued, individual plates being difficult to distinguish. Body colour, when wax removed, flesh pink, becoming grey rapidly after death. *Antennae* (fig. 20) 8-jointed. Basal joint broader than long, with 3–4 setae subapically on the distal edge. Second joint longer than broad. Third joint a little shorter than second. Fourth joint shortest, just over half length of second. Fifth joint same length, or slightly shorter than third. Sixth joint same length as fifth. Seventh joint same length as third. Eighth joint twice as long as third. Joints 2 to 7 with subapical rings of 4–5 setae, 7th joint also with one long, blunt, curved, subapical spine. Eighth joint with subbasal, median and subapical rings of setae, and 1 median and 3 subapical spines. Relative measurements of the joints, in microns, are: (1) 41; (2) 40; (3) 36; (4) 22; (5) 30; (6) 30; (7) 37; (8) 75.



Figs. 20–27.—*Farinococcus loranthi*, sp. n.: 20, antenna; 21, hind leg; 22, anal cerarius; 23, anal ring; 24, dorsal osteole; 25, dermal pores; 26, section of ventral abdominal dermis; 27, adult female.

Hind leg (fig. 21) reduced in size, short, stout. Coxa twice as broad as long, with a few setae medially and on the integumental edge and (hind pair only) numerous translucent pores on the outer face. Trochanter large and robust, with 3–4 setae, mostly on the distal edge. Femur nearly twice as long as broad, with three rows of setae, one medially, and one on either edge (usually 8–9 setae on the inner edge, and only 2–3 on outer edge). Tibia nearly twice as long as broad, with the usual three rows of setae. Tarsus twice as long as broad, with three rows of setae. Tarsal digitules thin and fine, with minutely knobbed ends; equal to tarsus in length.

Claw robust, sharply curved, half as long as tarsus. Ungual digitules ¾ as long as claw, thicker than tarsals, with more pronounced knobs at ends. Relative measurements of the legs, in microns, are given below :—

	Coxa		Femur		Tibia		Tarsus		Claw
	Long	Broad	Long	Broad	Long	Broad	Long	Broad	
Leg 1 ...	64	165	167	95	86	48	65	36	34
Leg 2 ...	64	167	163	93	84	42	68	32	38
Leg 3 ...	89	211	169	87	95	42	75	30	36

The third pair of legs are slightly longer than the other two pairs. *Cerarii* (fig. 22) nine pairs distinguishable on the abdominal 2nd and 3rd thoracic segments, 1 pair on the 1st thoracic segment, then either one or two poorly defined pairs forming, with the pair on the 1st thoracic segment, a practically continuous spine and pore band encircling the anterior segments. Spines long, narrow at base, often tapering to a thread-like point, interspersed with stoutish hairs and circular pores. Spines more or less arranged in 3–4 rows posteriorly, and 2–3 rows anterolaterally, with the anterior and inter-antennal ones clustered in no particular arrangement, and altogether forming a marginal fringe to each segment. Anal cerarius with one long seta in addition to the short, stoutish ones, and with a distinct sclerotised plate on the ventral aspect. On the ventral side of the cerarii there are numerous long setae, interspersed with circular, and apparently some oblong, pores. Spine counts of 5 adult specimens, commencing from the anal extremity were as follows : (1) 15–25, mean : 17 ; (2) 22–38, mean : 31 ; (3) 22–36, mean : 30 ; (4) 23–38, mean : 32 ; (5) 19–38, mean : 31 ; (6) 24–36, mean : 32 ; (7) 27–40, mean : 35 ; (8) 35–43, mean : 39 ; (9) 41–63, mean : 55 ; (10, 11 and 12) 112–199, mean : 183. Beak dimerous, second joint long, and acutely pointed. Measurements : 200 by 114 microns. Anal ring (fig. 23) with 6 setae and two rows of pores, the outer row roughly rectangular in shape, the inner row slightly larger, and circular in shape ; surrounded by an area clustered with circular pores and setae. *Dorsal osteoles* (fig. 24) small in size, but with thickly sclerotised lips with groups of pores and curved derm spines on outer edges. Eyes large and prominent, stout, thumb-like. Approximately the same size as the second antennal joint. *Derm* densely covered dorsally with pores of two types, numerous hair-like setae, and clusters of sharp, stout, curved spines, mostly on the mid-dorsum of segments. Derm pores either circular, with thickened lips, and a granulate lumen (commonest), or larger, oblong, with clear lumen. There are also a few large circular disc pores on the dorsum. It is likely that the oblong pores are, in reality, the same as these circular pores, but have become compressed during mounting. If so, the same fault has developed in the twelve specimens, mounted on seven different slides, examined. The small circular pores usually surround the oblong pores, which are more or less restricted to clusters mid-dorsally, and laterally dorsal to the cerarii (figs. 25 a & b). Ventrally with no spines and rather fewer pores and setae, though oblong pores also present, especially on the undersides of the cerarii, where long setae are also profuse. On the anterior three of the last four abdominal segments are clusters, arranged more or less in lines, of large 12-locular pores, with glassy outer rings and thickly demarcated loculi. The same segments also have groups of 8–10 tubular ducts in the distal corners of the margins (figs. 25 c & 26). There is one oval translucent area on the ventral aspect of the abdomen. The whole derm, dorsally and ventrally, is striated with faint zig-zag markings, giving, with the pores and setae, a " pepper-and-salt " appearance to the insect when viewed under the low power of the microscope.

Length 2·7 mm., breadth 1·9 mm.

Type Locality : Tafo, Gold Coast.

Type Host : *Loranthus bangwensis*, in domatium inside canker, attended by an undetermined ant of the genus *Crematogaster*. The *Loranthus* was epiphytic on *Discoglypremna caloneura*.

Material Examined : Twelve adult females. The type has been deposited in the British Museum collection.

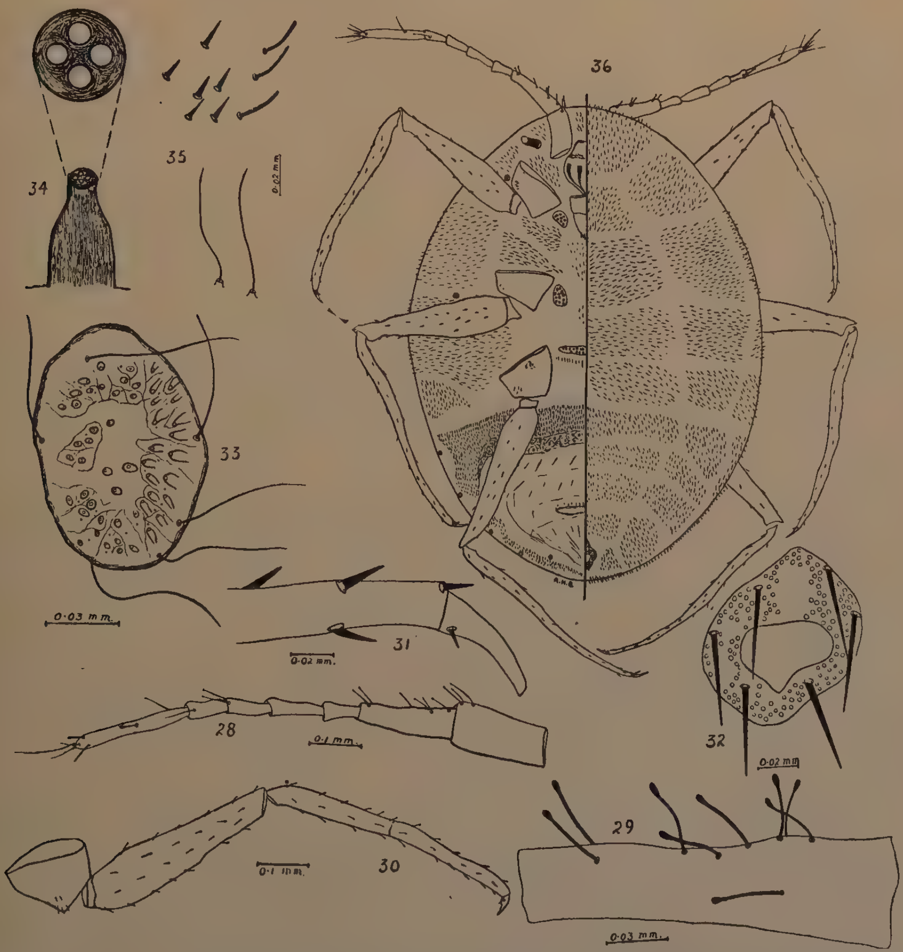
This species has been assigned to *Farinococcus*, Morrison (1922), on the grounds of the continuous lateral spinal band, and the large number of 12-locular pores in the genital region. It differs from the genotype, *F. multispinosus*, Morr., and from *F. formicarius*, Green, in possessing the three series of tubular ducts on the penultimate, and two preceding, abdominal segments ; in this respect, it comes near to *Paraputo*, Laing, to which it is undoubtedly closely related. It differs, however, in the conspicuous development of the cerarii, and in the 8-jointed antennae. *F. loranthi* would seem to occur in 75 per cent. of the *Loranthus* in the Tafo, Aburi, and Kumasi regions examined by the writer. It has been taken from the epiphytes growing on cacao, coffee, and several indigenous trees, and once, by D. Westwood, as a colony on a cacao pod, at Mampong, Eastern Province. In all, three species of *Loranthus* have been found infested, *L. bangwensis*, *L.* nr. *buntingii*, and *L.* nr. *micrantherus*, and three species of *Crematogaster* have been taken attending the mealybug, but there appears to be no correlation between the ant and mistletoe species.

(41). *Newsteadia wacri*, **sp. n.** (Figs. 28–36).

Adult Female (fig. 36) completely covered with white wax in the form of plates of a definite pattern. One large triangular plate covering the head and apex of the body, overlaid by a second, and smaller, plate on the first thoracic segment. Two pairs of triangular plates on the succeeding thoracic segments. Two wide plates on the anterior abdominal segments, and one large triangular plate covering the terminal abdominal segments. Six to seven pairs of plates situated laterally, the anterior three or four broadly triangular in shape, the posterior ones more finger-like, the anal pair being gently curved backwards, and overlapping the edges of the ovisac in mature specimens. Ovisac dorsally of six to seven wax strips cemented together to form a slightly curved, striated, plate ; ventrally, a trough of wax, evidently secreted entire. Body colour, when wax removed, very pale ochreous yellow. *Antennae* (figs. 28 & 29) 7-jointed, very much broader at base than at apex. Basal joint robust, half as broad as long, with a cluster of 2–3 stout, conspicuously knobbed, setae at the inner apex. Second joint broad at base, tapering to apex, with four groups of knobbed setae, one basally, two sub-medially, and one apically placed. Third joint one third length of second. Fourth joint half as long as second. Fifth joint equal in length to fourth, and usually with one pair of setae subapically. Sixth joint slightly shorter than fourth, with one long seta on inner apical edge. Seventh joint rather longer than second, thin, rounded at apex, with two setae at tip, one being a third longer, and stouter, than the other, and with four or five other, knobbed, setae distributed subapically, and one medially. Relative measurements of the joints, in microns, are : (1) 233 ; (2) 247 ; (3) 89 ; (4) 118 ; (5) 110 ; (6) 100 ; (7) 281. *Hind leg* (figs. 30 & 31) long and thin. Coxa broader than long, with a cluster of setae on the distal edge. Trochanter reduced, twice as broad as long, incompletely divided off from proximal end of femur. Femur rugged in outline, broad proximally, tapering at distal end, with four rows of knobbed setae, two medially, and one at either edge. Tibia fused to tarsus, slightly curved inwards, and twice as broad proximally as distally ; with three rows of knobbed setae. The point of fusion with the tarsus can be distinguished with some difficulty, the two joints being almost equal in length. Tarsus proximally with knobbed setae, distally with three rows of short, stout, sharp, spines. Claw gently curved, with digitules consisting of a pair of short, sharp, spines. Relative measurements of the joints, in microns, are given below.

	Coxa	Femur	Tibia-tarsus	Claw
Leg 1	172	674	694	52
Leg 2	189	715	897	62
Leg 3	203	754	975	62

Beak obscurely dimerous, short, stout, triangular, with a cluster of setae at apex, one seta on each half of the beak being very long, the rest short, and forming a tuft at the tip. Length 0·2 mm. Anal ring (fig. 32) with 6 stout, robust, sharply pointed setae, and two rows of more or less circular pores, though these are often difficult to distinguish as distinct rings. With two pairs of thoracic spiracles, relatively large, with 6–8 multilocular pores scattered around the openings. Five pairs of abdominal spiracles, very much smaller than the thoracic ones, and often hard to distinguish. One specimen had only 4 abdominal spiracles on one side.



Figs. 28–36.—*Newsteadia wacri*, sp. n. : 28, antenna ; 29, 2nd antennal joint ; 30, leg ; 31, end of tarsus and claw ; 32, anal ring ; 33, ventral pore cluster ; 34, pore from ventral cluster ; 35, derm spines and setae ; 36, adult female.

Genital orifice large and conspicuous, with a row of setae along the anterior lip. Eyes long, stout, thumb-like. Measurements 0·01 by 0·043 mm. *Derm* with numerous pads of spines dorsally and laterally, situated more or less in agreement with the wax plates, and ventrally between the legs, and anterior to the ovisac band. Median venter free of spines, but with two or three clusters of long, thin, setae. A few setae distributed throughout the spine pads, especially laterally. Spines less numerous ventrally. Between the coxae of the first two pairs of legs are two pairs of saucer-shaped clusters of papillate quadrilocular pores. Between the coxae of the hind pair of legs there is a single elongate oblong cluster of these peculiar pores. Around the edges of the pore clusters, there are usually 6–7 long, thin, setae (figs. 33 & 34). Ovisac band densely packed with curved spines and multi- (usually quadri-) locular pores separated by faint, but distinct, striations. Band not reaching quite to apex of abdomen, but tapering off just before reaching anus. Derm pores relatively sparse.

Length 2·0 mm., breadth 1·5 mm. as mounted on a slide. Living insect 2·2 by 2·0 mm. With ovisac 6·0 by 2·2 mm.

TYPE LOCALITY : Tafo, Eastern Province, Gold Coast.

TYPE HOST : *Theobroma cacao.* Coll. : *J. McA. Todd.*

MATERIAL EXAMINED : Eighteen adult females. Type deposited in the British Museum collection.

This species can be distinguished from all the other described species of *Newsteadia* by the peculiar papillate pore clusters on the mid-venter, and in particular from *N. floccosa*, Deg., by the 7-jointed antennae and general size, from *N. americana*, Morr., by the relative lengths of the antennal joints and the unconstricted nature of the dorsal abdominal spine bands, and from *N. mauritiana*, Mamet, by the 7-jointed antennae, and the spine-like ungual digitules.

This species was first collected feeding on the bole of a young cacao tree about two feet above ground level. One nymph was subsequently taken feeding on a rootlet, about 2 inches below the soil surface. It has subsequently been taken in very large numbers from the buttress roots of *Terminalia superba* and *Triplochiton scleroxylon* feeding under flakes of bark. All records from Tafo.

(42). *Icerya nigroareolata*, Newstead, 1917.

This large Monophlebid was first described from *Coffea* sp. and cotton at Kampala and Jinja, Uganda. It has been taken twice on cacao at Tafo, and once at Akwadum. Also taken at Tafo on *Hibiscus* sp., *H. rosa-sinensis*, *Ceiba pentandra*, *Corchorus acutangulus*, *Cissus* sp., *Psidium guajava*, *Rosa* sp., *Tephrosia* sp., *Acalypha* sp., *Codiaeum* sp. and on *Brownea* sp. at Bunsu (Eastern Province). By far the heaviest infestation was on *Acalypha* sp., and it seems as if this plant is the major host in the Tafo area. Mr. F. A. Squire brought back from Sierra Leone a mixed collection of this species with *I. maxima*, Newst., resulting in an extremely heavy infestation on a *Casuarina* sp. at Newton. Young nymphs of this species are almost invariably found clustered along the midribs on the undersides of the leaves of the host plant.

(43). *Steatococcus theobromae* (Newstead, 1908).

This species was originally described as *Paleococcus theobromae*, and, as such, was cited from cacao by von Faber (*l. c.*) from the Cameroons. It was described from cacao at Calabar. Morrison (1928) removed it to the genus *Steatococcus*, Ferris, where it now remains. Two collections have been examined by the writer, one from *Antiaris africana* at Tafo, the other from *Chrysophyllum* sp. at Owena, Nigeria. Both infestations were heavy, the insects being clustered mostly on the midribs on the undersides of the leaves, and on the young shoots. Cotterell has collected this species from cacao at Boma, Eastern Province.

(44). *Steatococcus* sp. (H. 5055).

This species, the identity being at present obscure, is, for the time being, referred to under its number in the W.A.C.R. Institute Coccid collection. It has been taken four times on cacao, all at Tafo, and all as single pre-adult specimens. It has also been taken at Tafo from a branch of *Sterculia tragacantha*, about a dozen young adults, some of which were covered with bright sulphur-yellow wax, some being covered with pure white wax (Coll. : *R. J. Benstead*), and once from Aburi, from *Acacia arabica*.

Non-African Cacao Coccids.

The species listed below have been taken from cacao in parts of the world other than West and Central Africa and San Thomé, with the reference from which the record was obtained.

Pseudoparlatoria pusilla, Green. Ceylon. Green (1922a).

Howardia biclavis (Comst.). Trinidad. Pickles (1941).

Asterolecanium pustulans (Ckll.). Trinidad. Pickles (1941).

Asterolecanium aureum (Bdv.). Dominican Republic. Gomez-Menor y Ortega (1934).

Ceroplastodes virescens, Green. Ceylon. Green (1909).

Philephedra broadwayi, Ckll. Trinidad. Green (1915).

Tachardia albizziae, Green. Ceylon. Green (1922).

Rhizoecus moruliferus, Green. Trinidad. Strickland (1945).

Morrisonella globocula, Hambl. Trinidad. Hambleton (1946).

Morrisonella theobromae, Hambl. Ecuador. Hambleton (1946).

Ripersiella ornata, Hambl. Trinidad. Hambleton (1946).

Pseudococcus theobromae (Dougl.). England. Douglas (1889).

Pseudococcus tayabanus, Ckll. Philippines. Cockerell (1905).

Pseudococcus lilacinus, Ckll. Ceylon. Green (1922).

Pseudococcus bromeliae (Bch.). Barbados. Ballou (1922).

Ortheziopa sp. Trinidad. Strickland (1945). (*Note.* This species has recently been identified by Dr. Hall as *O. reynei*, Laing.)

Lophococcus convexus, Morr. Dutch East Indies. DeJong (1931).

Acknowledgements.

The writer wishes herewith to acknowledge his great indebtedness to Dr. W. J. Hall, Director of the Imperial Institute of Entomology, without whose constant help and encouragement this paper would never have been written. Dr. Hall has, in fact, identified, or confirmed, the identity of all the species of Coccids collected by the writer, and detailed in the present paper. Acknowledgements are also due to Mr. N. D. Riley, Keeper of Entomology at the British Museum (Natural History), for allowing the writer to work for a time in the Department of Entomology, to Mr. F. Laing for making available the Coccid collection at the British Museum and allowing the writer to examine his host plant and locality records, to Dr. V. B. Wigglesworth for allowing the writer to carry out certain parts of the systematic work at the Entomological Laboratory at Cambridge, to Messrs. J. Paine and D. Westwood, Agricultural Officers, Gold Coast Department of Agriculture, for much material collected by them whilst on tour in various parts of the Colony, and especially to Mr. J. McA. Todd, Pathologist, W. African Cacao Research Institute, for identifying many of the host plants listed.

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A RECORD OF *SYNTOMOSPHYRUM GLOSSINAE* FROM NIGERIA.

By T. A. M. Nash, O.B.E., D.Sc.,
Medical Entomologist, Sleeping Sickness Service, Nigeria.

In April 1947, the laboratory stock of *Glossina palpalis*, R.-D., puparia collected from the Kaduna area suddenly became heavily infested with the pupal parasite, *Syntomosphyrum glossinae*, Wtstn. Presumably an infected pupa had been brought in from the bush and a few parasites had emerged undetected ; it was not until the emergence of their offspring that the mass-infestation was revealed.

This is believed to be the first record from West Africa of this parasite of tsetse pupae. Mr. Nixon, who kindly confirmed my identification, informs me that there are no specimens from West Africa in the British Museum.

It should be mentioned that twenty-three years ago Lloyd and Johnson (Bull. ent. Res., 17 p. 449, 1927) bred large numbers of this parasite from Nyasaland stock. 1/ Adults and infected pupae were liberated in the breeding sites of *G. morsitans*, Westw., and *G. tachinoides*, Westw., at Sherifuri, which is 210 miles north-east of Kaduna. No tsetse pupae subsequently collected from the site were parasitised by this insect, nor were thousands of pupae collected in the adjacent country between 1927 and 1943. (Possibly the very high dry season temperature and low humidity at Sherifuri were responsible for the complete failure of the experiment.)

Undoubtedly the recent Kaduna record indicates that *S. glossinae* is indigenous in Nigeria ; it cannot be attributed to Lloyd and Johnson's abortive efforts of twenty-three years ago.

STUDIES ON *ANOPHELES GAMBIAE* AND *A. MELAS* IN AND AROUND LAGOS.

By R. C. Muirhead Thomson, D.Sc.
Colonial Medical Research.

X.6.

(Plate X.)

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The Identity of *A. gambiae* **and** *A. melas.*

In the last few years the identity of "*A. gambiae* var. *melas*" Theo., has become much clearer. Ribbands (1944) on the basis of salinity reactions of the larvae, characters of the larval pecten, and maxillary palp banding of the adults, considered it was a separate species. The specific status was supported by Muirhead Thomson (1945) on the basis of consistent and well-marked differences between the eggs of the two species. Chwatt (1945), however, working in a different part of West Africa—Lagos—considered that none of these characters was reliable enough to justify specific status for *A. melas.*

In the present investigation one of the first questions to be settled was the identity of *A. melas* and *A. gambiae*, Giles, with particular reference to the adults. In Freetown Estuary, Sierra Leone, about one-third of the *melas* adults had an additional dark band on the palps by which they could be distinguished from *A. gambiae* adults, in which the extra dark band only appears rarely. The remaining two-thirds of the *melas* had no extra dark band, and could only be distinguished from *A. gambiae* by egg characters.

Previous reports had shown that in the Gold Coast and Nigeria the proportion of *melas* that was 4-banded was much smaller than in Sierra Leone. In Lagos only about 5 per cent. of the *melas* were found to have 4-banded palps, the remaining 95 per cent. being indistinguishable morphologically from *gambiae.* There remained the egg character as a means of identification.

In Sierra Leone, an examination of over 1,000 batches of eggs laid by wild caught females, and about 3,000 individual eggs collected from breeding places, showed the egg characters to be distinct and completely reliable.

In Lagos, Chwatt (1945) had found that 92 per cent. of the eggs could be assigned to *melas* or *gambiae* forms, but that 8 per cent. could not be classified.

In the present investigation over 1,200 egg batches were examined from wild caught females; only 4 of these batches presented any difficulty, the remaining 99·7 per cent. of the eggs could be distinguished at once with no difficulty, and the character remains, therefore, for all practical purposes, an absolutely reliable diagnostic character between *A. melas* and *A. gambiae.* The difference can be immediately detected in unhatched eggs on filter paper, examined under the low-power binocular microscope. Under these conditions African assistants can be trained in a short time to distinguish the two types; on checking the results the character evidently presents no difficulties to them. Resort to the use of the high-power monocular microscope, and to careful measurement, is quite unnecessary, and actually makes distinction much more difficult. Similarly, hatched eggs with collapsed chorion cannot be identified with any accuracy. It seems likely that the use of one or all of those methods explains the difficulties some observers have found with this character, and their adoption merely tends to confuse the issue.

The character of the egg remains so far the only reliable feature distinguishing all adult female *melas* from *gambiae.* In Lagos, where the proportion of 4-banded *melas* is so small, the egg character assumes an even greater importance than in Sierra Leone, and it is quite impossible to form even a rough estimate of the relative proportion of adult *melas* and *gambiae* in a collection without inducing a sample of females to lay eggs.

Identification of the eggs forms the basis of work described below on the seasonal incidence of *gambiae* and *melas* in houses, and on the seasonal infectivity of the two species.

Further proof of the specific status of *melas*, described later in this paper, shows that adults bred from *melas* eggs can be induced to mate in the laboratory, and that they subsequently all lay distinct *melas* form eggs. Similarly with *gambiae,* the egg form remains true to type.

Finally, by cross fertilisation experiments it is shown that *melas* cross mated with *gambiae* can produce a healthy generation of hybrid adults. Of these hybrids the females appear to be normal sexually, but the males from both crossings show various degrees of abnormality in the testes, and are evidently sterile. The mutual sterility of hybrids is a severe test of specific differences, and considered in conjunction with clear-cut egg characters, and with the physiological and morphological differences between the larvae, it forms a reasonably sound basis of fact on which the specific status of *A. melas* in West Africa is established.

Rainfall.

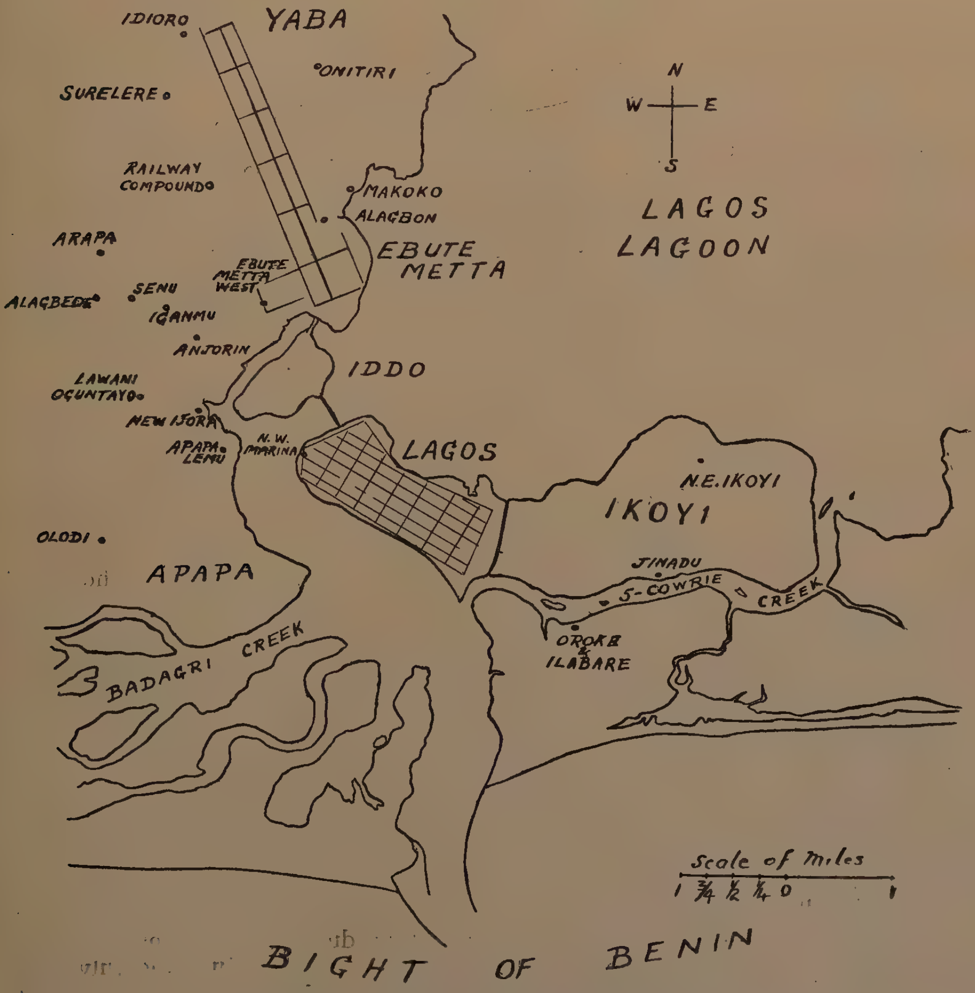
The general meteorological conditions in the Lagos area have been described by Barber and Olinger (1931). The distribution and intensity of rainfall may vary considerably from year to year, and these changes undoubtedly determine the variations in seasonal incidence of *gambiae* from place to place, and from year to year.

In general there is a well-marked dry season during the months of December, January and February. The season of heavy rains is in May, June and July. August is usually a month of low rainfall, and at that time of the year there may be actually a short dry season of 6 or 7 weeks followed by a minor wet season from about mid-September to about mid-November.

The distribution of rainfall from 1942 to 1944 is shown by Gilroy and Chwatt (1945), and that of 1945 and early 1946 in fig. 1 of the present paper.

During the course of the present investigation there were two noteworthy rainfall features which influenced the incidence of *gambiae* in different catching stations. Firstly, the occurrence of intermittent heavy showers in February and March 1945 produced a sharp increase in *gambiae* incidence two months before the real rainy season started. This feature was absent in 1946. Secondly, the little dry season of August 1945 was well marked and prolonged, and imposed a severe check on *gambiae* breeding in rain-filled pools and puddles.

As will be described in the following sections the effect of rainfall on *gambiae* incidence may vary considerably, even within the limited area shown in Map 1, depending on the nature of local breeding places.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OLODI
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5-COWRIE CREEK
BADAGRI CREEK
OROKE & ILABARE
Scale of miles
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BIGHT OF BENIN

Map 1. Lagos and environs showing catching stations.

Seasonal Incidence of *A. gambiae* and *A. melas* in Houses in the Lagos Area.

In Sierra Leone the proportion of adult *melas* which have an additional dark band on the palp is sufficiently high to enable rough estimates to be made of its seasonal abundance in houses. In the Lagos district of Nigeria about 95 per cent. of the *melas* have unbanded palps, and are indistinguishable morphologically from *gambiae*.

Fortunately the egg distinction remains clear and reliable, and affords an accurate, if rather laborious, method of distinguishing the two species.

At the beginning of this investigation there was no information available about the incidence of these two species in the Lagos area, and it therefore seemed of first importance to establish as soon as possible how abundant the two species were in different localities, and how their proportions varied at different seasons.

Catching stations were selected at different places in and around Lagos in African native houses. As all the mosquitos were required alive and in good condition for laying eggs, hand-catching by the suction tube was the method selected. Whatever advantages or disadvantages hand-catching has compared with the spray sheet method in checking up on Anopheline incidence, it is clearly the only feasible method here. Spraying may yield a higher catch in a house, but the dead material cannot be identified, and there is no way of telling whether *gambiae* or *melas* predominates in the catch ; without this distinction the whole point of routine house catching is lost.

The number of bedrooms searched varied according to the abundance of *Anopheles*. Sometimes two or three rooms supplied enough material, but 10–20 rooms was a more usual range. On returning to the laboratory a good sample of the catch—anything up to 50 females—was taken, and each mosquito isolated in a 6×6×6 inch cage. A bowl of water was provided for egg laying, and blades of grass scattered on the surface to facilitate oviposition. In this way it was ensured that the maximum number of mosquitos not only laid eggs, but survived in good condition for later dissection. Oviposition takes place over two or three days after catching, and from the eggs

TABLE I.

Alagbon.—Seasonal incidence of *melas* and *gambiae* in houses.

Month	Number of visits.	Total *Anopheles*	Egg batches		*Anopheles* per 10 bedrooms	
			melas	*gambiae*	*melas*	*gambiae*
1945—Jan.	1	24	22	1	32	2
Feb.	9	340	125	11	45	4
March	11	725	232	119	60	34
April	8	285	62	102	22	31
May	4	125	9	51	7	45
June	5	261	10	71	10	66
July	2	132	19	27	38	56
Aug.	3	92	4	28	5	45
Sept.	3	23	7	1	14	1
Oct.	4	67	25	13	17	10
Nov.	2	19	6	4	10	6
Dec.	2	10	3	0	10	0
1946—Jan.	2	6	2	1	3	1
Feb.	3	62	14	4	17	12
March	1	3	2	0	5	0
April	1	7	4	0	14	0
May	1	66	17	17	66	66
June	1	42	3	15	7	35
	63	2,289	566	465		

identified the proportion of *melas* and *gambiae* in the catch was worked out. Fortunately, both species oviposited equally readily in the laboratory. The catch was standardised to an arbitrary unit of the number of female *gambiae* or *melas* per 10 bedrooms. It was originally intended to collect in about 10 catching stations every week or at least every fortnight right through the season, but pressure of other work made such frequent visits possible in only a few cases.

In considering these figures for room densities at different times of the year, the figures can be compared with other catches in the same catching station, but not with room densities in other localities. In a crowded urban area it may need a huge population of mosquitos to give a catch of one mosquito per room. Whereas in an isolated house the same population may give a catch of several hundred.

TABLE II.

Makoko.—Seasonal incidence of *melas* and *gambiae* in houses.

Month	Number of visits	Total *Anopheles*	Egg batches		*Anopheles* per 10 bedrooms	
			melas	*gambiae*	*melas*	*gambiae*
1945—Feb.	2	138	56	0	85	0
March	2	104	—	—	—	—
April	4	128	51	18	24	9
May	2	148	9	60	13	133
June	10	645	74	164	96	215
July	7	399	69	123	46	96
Aug.	7	348	66	55	45	34
Sept.	4	197	97	10	75	8
Oct.	2	27	13	2	43	6
Nov.	2	44	27	16	28	16
Dec.	2	18	8	2	13	5
1946—Jan.	2	62	32	1	43	2
Feb.	3	156	66	1	58	1
March	5	109	27	0	16	0
April	2	57	26	0	14	0
	56	2,580	621	452		

The results of all these catches are set out in Tables I–VIII and fig. 1. The tables include the number of identified egg batches on which these figures are based. Only in the case of Lagos town was it considered necessary to include the number of rooms searched, as that was the only locality where several months yielded negative catches, in which case one naturally wants to know how many rooms were examined.

The catching stations, and the seasonal fluctuations in the number of *gambiae* and *melas* which were recorded, will now be considered separately.

For the present purposes the Lagos area can be divided into three zones (Map 1):

(1) The Mainland. The coastal strip from Yaba to Apapa.

(2) Lagos town on Lagos Island.

(3) Ikoyi and the 5-Cowrie Creek villages.

There were no catching stations on Iddo Island, or on the three reclaimed points of land (Magazine, Middle, and Meridian Point) across the water from Lagos.

Alagbon.

A little brick-field village of 7 or 8 houses on an island surrounded by brackish swamps (*Paspalum*, *Avicennia*, and *Rhizophora*) just east of Ebute Metta. In the early months of 1945—in the dry season—*melas* was dominant in the houses, and remained so till the middle of March. Then there was a sharp increase in the

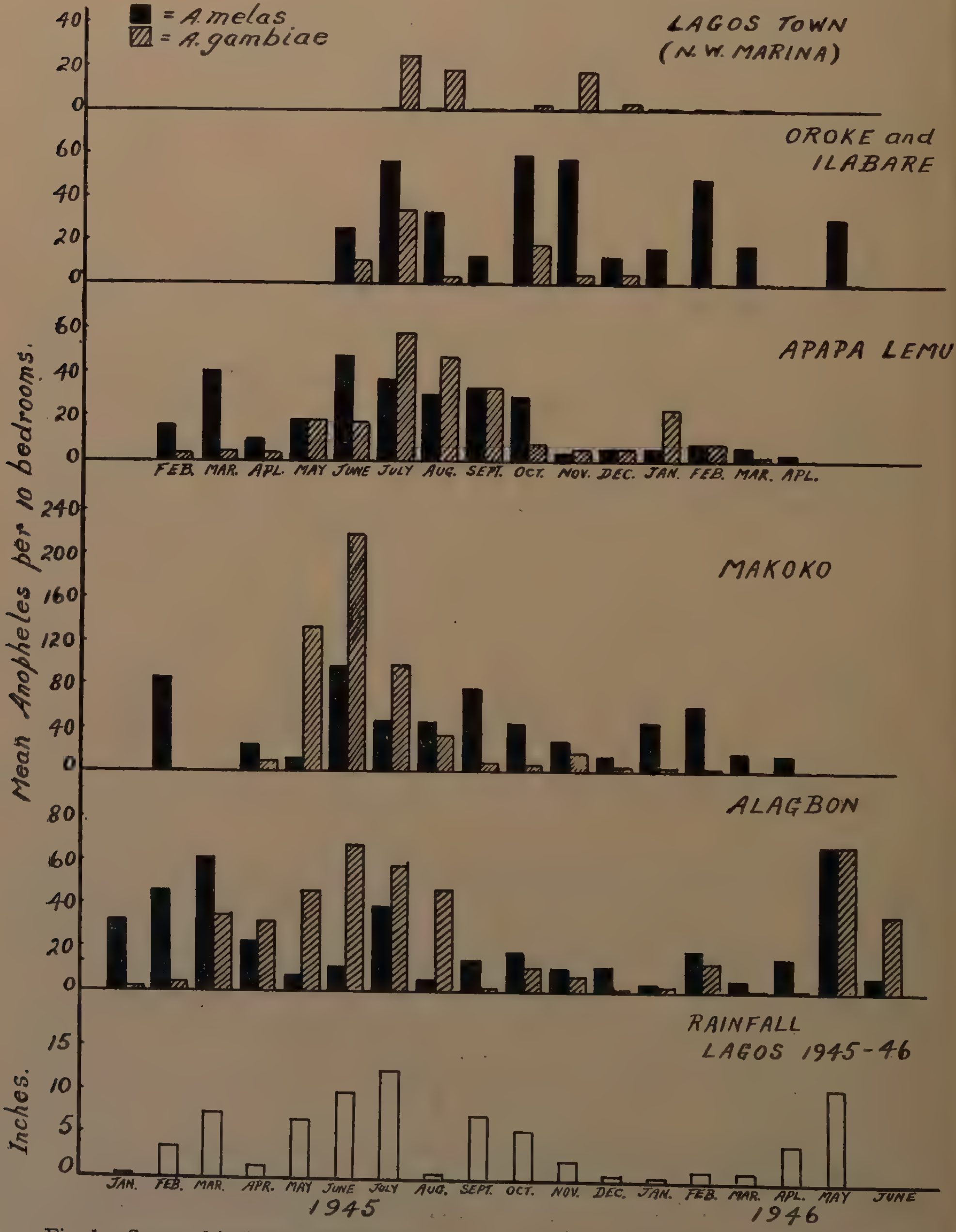

Fig. 1.—Seasonal incidence of *A. melas* and *A. gambiae* in houses from five localities in Lagos area 1945–46.

numbers of *gambiae*, which remained the dominant mosquito till August. As can be seen from fig. 1 this sharp increase in *gambiae* incidence occurs about two months before the proper rainy season, and was evidently related to the odd heavy showers in late February and early March. In 1946 the *gambiae* increase did not take place till May.

From September to April *melas* is dominant, but in some months the numbers are surprisingly low considering the great extent of brackish swampland surrounding the village.

Makoko.

An isolated village of about 60 houses on an island surrounded by brackish swamps one-third of a mile east of Ebute Metta. *Melas* is abundant throughout the year, and is the dominant mosquito except during the three rainiest months. In May, June and July, there was an enormous production of *gambiae*, undoubtedly caused by dense breeding in a few rain-filled borrow pits beside the village. As in Alagbon, the short dry period in August imposes a severe check on *gambiae*, and it disappears completely by the end of the dry season.

Onitiri.

An isolated brick-field village surrounded by extensive *Paspalum* and *Rhizophora*, half-mile east of Yaba. A few collections were made here in February and March 1945, at a time when large numbers of *melas* were being recorded at Makoko at the lower part of the brackish swamp. In that short time *gambiae*, surprisingly, was the dominant mosquito, and *melas* was not very common considering the situation of the village surrounded by hundreds of acres of tidal swamp. The *gambiae* dominance was apparently due to a single borrow pit, which retained water throughout the dry weather ; when this was treated with paris green, only odd *gambiae* were taken in the village.

Surelere.

This is a growing suburb, just west of Yaba. The catches here are interesting in that *melas* has not been found. The village is a little over a mile from the great stretch of tidal swamp east of Yaba and Ebute Metta, and little over 1½ miles from Makoko village where high catches of *melas* were regularly recorded. With the Ebute Metta—Yaba suburbs acting as a barrier between Surelere and the breeding grounds, it appears as if *melas* has little tendency to move inland any distance from the narrow coastal fringe.

Ebute Metta (West).

Irregular catches in this suburb show that both *melas* and *gambiae* occur in houses here. Unfortunately no catches were made in the rainy season when there was extensive breeding of *gambiae* in rain-filled puddles among the houses ; at that time *gambiae* is almost certainly the dominant mosquito in houses.

Iganmu, Alagbede, and Senu.

These are small villages on a narrow strip of dry land between a tidal creek on the one hand, and an extensive fresh water swamp or marsh on the other. *Gambiae* is dominant here, but *melas* occurs in numbers at certain times of the year. This is one of the few catching stations where *gambiae* is abundant right through the dry season and there is evidently steady production of this species from the fresh water swamp.

Railway Compound.

High catches were recorded in European bungalows and in servant quarters in March and April 1945, over 90 per cent. of the catches being *gambiae*. These unusually high catches coincided with very high figures for *gambiae* in the villages of Alagbon, three-quarter's of a mile to the east, and Alagbede and Senu, about one mile to the west.

Apapa Lemu.

This was a large isolated village of several hundred houses on a peninsula of land surrounded by mangrove and tidal swamp. Both *melas* and *gambiae* are abundant for most of the year, and the seasonal incidence of *gambiae* in particular is quite different from that of the catching stations above. The figures for *gambiae* are related to the peculiar nature of the breeding-place, which was a limited area of about 20–30 yards square of sedge swamp forming a narrow fringe between the upper edge of the mangrove belt and the dry land. This limited area was subject to drying on the one hand, and being flooded by the highest spring tides on the other.

TABLE III.

Apapa Lemu.—Seasonal incidence of *melas* and *gambiae* in houses.

Month	Number of visits	Total *Anopheles*	Egg batches		*Anopheles* per 10 bedrooms	
			melas	*gambiae*	*melas*	*gambiae*
1945—Feb.	2	62	25	3	15	2
March	2	98	60	8	40	4
April	2	29	18	6	9	3
May	1	56	17	17	18	18
June	4	389	82	30	47	17
July	7	757	140	188	37	58
Aug.	8	902	137	214	30	47
Sept.	11	1,170	341	353	33	33
Oct.	8	750	300	76	29	7
Nov.	4	112	22	31	3	4
Dec.	10	416	101	94	5	5
1946—Jan.	7	686	78	307	5	23
Feb.	5	296	54	94	8	8
March	4	175	50	22	6	2
April	4	44	16	2	3	0·5
	79	5,942	1,441	1,445		

There is no sharp increase in *gambiae* incidence in March and April—such as occurred in Alagbon—because high tides frequently flooded the breeding grounds. Production of *gambiae* remained high during the rainy season not only on account of the rainfall, but also because for several months no spring tides reached this patch of sedge. The little dry season of August did not produce a sharp fall in the numbers of *gambiae*, because rain-filled pools and puddles which dry quickly were not the source of the *gambiae* in this area.

The sharp fall in October was caused by exceptionally high spring tides reaching the breeding ground. These high spring tides kept *gambiae* breeding low for three months, but they also prevented the edge of the swamp from drying up in the dry weather, and as a result there was an increase in *gambiae* incidence in January—a very dry month, when *gambiae* breeding is low in most other catching stations.

Olodi.

This large village west of Apapa could only be visited once a month. It is situated on sandy soil near extensive mangrove and *Paspalum* swamps. There are a large number of shallow earth wells holding water throughout the year, but it is evident that heavy *gambiae* breeding is dependent on heavy rainfall from April to July.

TABLE IV.

Olodi.—Seasonal incidence of *melas* and *gambiae* in houses.

Month	Number of visits	Total *Anopheles*	Egg batches		*Anopheles* per 10 bedrooms	
			melas	*gambiae*	*melas*	*gambiae*
1945—March	2	121	55	6	47	3
April	1	52	1	28	2	50
May	1	52	5	16	7	22
June	1	92	19	8	43	18
July	1	145	12	32	63	178
Aug.	1	38	9	3	24	8
Sept.	1	23	10	2	12	3
Oct.	1	75	38	8	25	5
Nov.	1	24	7	1	14	2
Dec.	1	6	0	1	0	1
1946—Jan.	—	—	—	—	—	—
Feb.	1	8	3	2	1	1
March	1	53	6	1	27	4
	13	689	165	108		

Lagos Town.

The dense town of Lagos proper, on Lagos island, offers an interesting contrast to the catching stations described so far. Except at the extreme east end of the island, near the canal, there are no mangrove or brackish swamps, and no obvious breeding places for *melas*.

TABLE V.

New Ijora.—Seasonal incidence of *melas* and *gambiae* in houses.

Month	Number of visits	Total *Anopheles*	Egg batches		*Anopheles* per 10 bedrooms	
			melas	*gambiae*	*melas*	*gambiae*
1945—Dec.	3	108	42	2	25	2
1946—Jan.	9	293	87	58	14	9
Feb.	2	33	11	6	6	3
March	1	23	14	0	14	0
April	1	35	4	9	6	12
	16	492	158	75		

The part of the town which was of the greatest interest, as it was to Barber in his 1931 survey, was the north-west part of the island projecting into the lagoon directly opposite Apapa Lemu village. Barber found a higher incidence of "*gambiae*" in houses in this part than in any other part of the dense township, and suggested that invasion from across the lagoon accounted for this. At that time, of course, there was no means of telling whether the Lagos "*gambiae*" were *gambiae* or *melas*.

In the present investigation several collections in the early part of 1945 yielded nothing in these houses, so collecting in earnest was not taken up till July, at a time when the incidence of *gambiae* was at its height. Most of the collecting was done in small dark huts and hovels in and around Johnson Street, N.W. Marina. The results are shown in Table VI. Of 279 adults collected between July 1945 and March 1946, 96 per cent. were *gambiae*, and only 4 per cent. were *melas*. During most of that time there was continuous heavy production of *melas* just three-quarters of a mile across the lagoon as shown by the catches at Apapa Lemu, high catches of *melas* being recorded in months such as September and October when not a single *melas* was found in the N.W. Lagos catch. As these negative catches in Lagos are based on a large number of suitable rooms searched, it appears fairly certain that, whatever may have happened in the past, there is at present very little indication of *melas* invading Lagos from the mainland across the water.

TABLE VI.

Lagos (Johnson St., N.W. Marina).—Seasonal incidence of *melas* and *gambiae* in houses.

Month	Number of visits	Rooms searched	Total *Anopheles*	Egg batches		*Anopheles* per 10 bedrooms	
				melas	*gambiae*	*melas*	*gambiae*
1945—July	2	16	41	1	25	1	25
Aug.	5	54	107	3	49	1	18
Sept.	3	42	0	0	0	0	0
Oct.	3	39	7	0	3	0	2
Nov.	5	68	99	0	51	0	17
Dec.	6	83	25	2	12	0·3	3
1946—Jan.	3	128	0	0	0	0	0
Feb.	1	50	0	0	0	0	0
March	1	52	0	0	0	0	0
	29		279	6	140		

As for *gambiae*, the incidence of which in N.W. Lagos is closely dependent on rainfall, it will be shown in a later section on breeding habits that the nature of local breeding places in the Johnson Street area is such as to account easily for all the *gambiae* found in houses, and that there is no need to postulate invasion of this species from the mainland.

Oroke and Ilabare.

These are two villages on the south side of 5-Cowrie creek, and fairly representative of conditions all along the creek. They are almost entirely surrounded by mangrove and great stretches of *Paspalum* swamp, as yet uncontrolled. Throughout the catching season there was a fairly steady production of *melas*, which is always dominant. The short *gambiae* season is again evidently determined by the rainy season.

TABLE VII.

Oroke and Ilabare (5-Cowrie Creek).—Seasonal incidence of *melas* and *gambiae* in houses.

Month	Number of visits	Total *Anopheles*	Egg batches		*Anopheles* per 10 bedrooms	
			melas	*gambiae*	*melas*	*gambiae*
1945—June	1	68	43	20	25	11
July	1	45	21	13	56	34
Aug.	1	27	20	3	33	4
Sept.	1	25	7	0	13	0
Oct.	1	116	43	13	59	18
Nov.	2	90	41	4	58	5
Dec.	4	109	38	16	13	5
1946—Jan.	4	114	58	0	17	0
Feb.	2	171	84	0	48	0
March	1	59	22	2	18	1
April	—	—	—	—	—	—
May	1	56	40	1	30	1
	19	880	417	72		

TABLE VIII.

Incidence of *melas* and *gambiae* in minor catching stations.

Locality	Number of visits	Number of bedrooms	Total *Anopheles*	Egg batches		*Anopheles* per 10 bedrooms	
				melas	*gambiae*	*melas*	*gambiae*
Surelere ...	9	193	81	0	32	0	4
Onitiri ...	3	35	218	24	86	18	51
Ebute Metta (West) ...	7	82	132	54	17	15	5
Railway Cmpd. ...	6	—	223	10	93	—	—
Ikoyi N.E.	8	65	236	86	8	31	4

Ikoyi.

Catches in the boys' huts in the European Reservation were made between February and July, 1945. Of 236 mosquitos caught, 90 per cent. were *melas* and 10 per cent. *gambiae*, the *gambiae* season being restricted to the two rainiest months of June and July. The collections were discontinued when house spraying with Pyrethrum, and later DDT, was started in the reservation. The dominance of *melas* in Ikoyi island is related to the vast area of tidal swamp—now reclaimed—round the island, and to the sandy nature of the soil which discourages freshwater pool formation even in the wettest months.

Summary of catching station results.—If the area bounded by Yaba, Apapa, and Ikoyi is taken, it is seen that *melas* and *gambiae* are about equally abundant in houses in this area as a whole. But if the individual catching stations are considered, wide differences are evident in the abundance of *melas* and *gambiae* in different localities.

Along the coastal fringe of the mainland *gambiae* and *melas* are about equally abundant, but at a very short distance from the coast, *melas* fades out. For example, it is still present in small numbers in Arapa and Railway Compound at the extreme upper end of tidal creeks, but has not yet been found in Surelere, a little over a mile inland from the coastal swamps.

On Lagos Island *gambiae* is the dominant mosquito, with only occasional *melas* recorded in the western half of the island.

In 5-Cowrie creek and Ikoyi Island *melas* is by far the most abundant mosquito in houses, and *gambiae* only appears in any numbers during the heavy rains.

Catching stations within a short distance of each other may have widely different mosquito populations. For example, for weeks on end the catch in Anjorin was almost entirely *melas*, while at Alagbede and Senu, little over half mile away across an open swamp, the catch was almost entirely *gambiae*.

Similarly, the small village of Lawani Oguntayo only yielded odd mosquitos at a time when high catches were taken regularly in New Ijora (½ mile) and Apapa Lemu (¾ mile). In 4 visits in January and February 1946 the total mosquitos caught in these three stations were, Apapa Lemu 276, New Ijora 84, Lawani Oguntayo 5.

Not only does the proportion of *melas* and *gambiae* vary greatly in different stations, but the incidence at different times of the year, also, does not necessarily follow the same trend throughout the area. With *melas*, the peak months vary from station to station, and are not necessarily related to rainfall. The abundance of *melas* is mainly determined by tidal movements, which are not only variable and unpredictable, but which even in this restricted lagoon area may have widely different effects in places a few miles apart.

The seasonal fluctuations in numbers of *melas* in houses can only be understood by studying local conditions of breeding which will be discussed in a later section.

The abundance of *gambiae* is much more obviously related to the rainfall, and in most stations the dry season brings about a great reduction in numbers. In localities like Apapa Lemu and Alagbede where the *gambiae* originates from swamp edges and not from rain-filled pools and puddles, the *gambiae* season is prolonged. Here again the incidence of adults can only be appreciated in the light of local breeding conditions.

A final point which emerges, even in this short period of 18 months, is that in any one catching station the seasonal incidence of *melas* and *gambiae* may follow an entirely different course in consecutive years. A glance at the chart will show that in Alagbon, for example, there is no resemblance between catches in the first 6 months of 1945, and the same period of 1946. The figures in this report, therefore, are only of limited use in forecasting the periods of peak abundance of these Anophelines ; they only show the range and variety of seasonal incidence that may be encountered.

The Natural Infectivity of *A. melas* and *A. gambiae* in the Lagos Area.

When this investigation started there was no information of any kind to show the separate parts played by *melas* and *gambiae* in malaria transmission in and around Lagos. As there were no clear distinguishing features between *melas* and *gambiae* at the time of Barber's survey in 1930, all his records of " *A. gambiae* " dissections from this coastal area refer to mixed populations of the two species in variable proportions.

TABLE IX.

Dissections of *melas* and *gambiae* from all catching stations in the Lagos area. June 1945–April 1946.

Locality	*melas*			*gambiae*		
	Number dissected	Glands positive	Sporozoite rate	Number dissected	Glands positive	Sporozoite rate
			Per cent.			Per cent.
Apapa Lemu	795	36	4·5	947	105	11·1
Makoko	313	12	3·8	183	18	9·8
Anjorin	322	1	0·3	63	1	1·6
Oroke and Ilabare ...	164	9	5·5	31	2	6·5
Makoko (outside) ...	113	3	2·7	29	2	6·9
Lagos Town	6	0	0·0	104	8	7·7
New Ijora	97	2	2·1	49	8	16·3
Other stations	129	4	3·1	149	12	8·1
Totals	1,939	67	3·5	1,555	156	10·0

This work clearly needed complete revision in the light of recent knowledge about the identity of *melas* and *gambiae*. Accordingly routine dissections were started in conjunction with the work described in the previous section on seasonal incidence of the two species in houses.

Only those adults which laid eggs in the laboratory and survived were dissected, and only salivary glands were examined for infections. Of the 3,494 *melas* and *gambiae* dissected, 2,132 were dissected by my assistant, Mr. Augustine Ameachi using Barber's method, and 1,362 were dissected by the author using Shute's technique. Material from all catching stations was dissected, but most attention was given to the large village of Apapa Lemu where abundant *melas* and *gambiae* occurred together in houses for many months of the year, and where there was a unique opportunity of studying these two species when they were exposed to chances of infection as equal as it is possible to find in nature.

The results from all stations are shown in Table IX.

All stations agree in showing consistently that *gambiae* has a much higher sporozoite rate than *melas*; the actual figures for all stations being 10·0 per cent. and 3·5 per cent., showing that *gambiae* is nearly three times as efficient a vector as *melas*.

The only other figures for *melas* in West Africa, where identifications were based on egg characters and palpal banding, are those of Tredre (quoted by Muirhead Thomson 1945) in which 1,000 gland dissections of material from Freetown Estuary gave a sporozoite rate of 4·2 per cent.

In the Lagos figures it will be seen that *melas* from Anjorin had an unusually low rate of infection, doubtless related to the fact that the catching station was a single isolated house beside extensive breeding grounds. If we exclude these catches we get a sporozoite rate of 4·1 per cent. for *melas* as more representative of conditions in the Lagos area, which agrees very closely with the Freetown Estuary, Sierra Leone, figure of 4·2 per cent. quoted above.

In Apapa Lemu the monthly changes in sporozoite rate of *melas* and *gambiae* caught in houses are shown in Table X, and these fluctuations are compared with the monthly incidence of the two species in the village houses in fig. 2.

TABLE X.

Gland infections of *melas* and *gambiae* in Apapa Lemu by months. June 1945–April 1946.

	melas			*gambiae*		
	Number dissected	Glands positive	Sporozoite rate	Number dissected	Glands positive	Sporozoite rate
			Per cent.			Per cent.
1945—June	46	2	4·3	15	3	20·0
July	76	1	1·3	172	2	1·2
August ...	95	3	3·1	167	21	12·5
September ...	223	18	8·0	250	29	11·6
October ...	169	7	4·1	49	14	28·6
November ...	13	0	0	15	3	20·0
December ...	67	0	0	49	8	16·3
1946—January	36	2	5·6	166	18	10·8
February ...	35	1	2·8	52	5	9·6
March and April	35	2	5·7	12	2	16·7
Totals	795	36	4·5	947	105	11·1

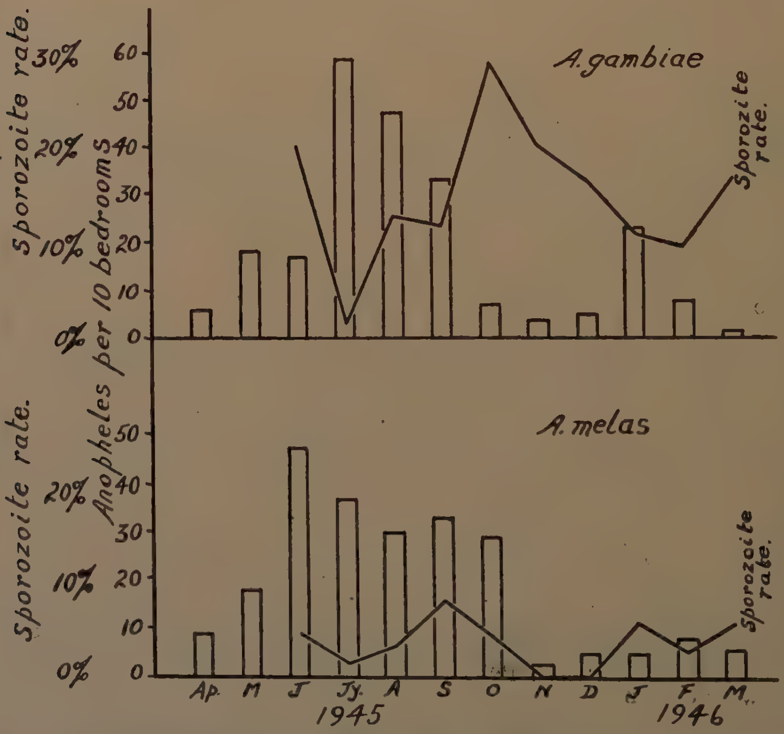

Fig. 2.—Seasonal incidence of *A. gambiae* and *A. melas* in houses at Apapa Lemu compared with seasonal infectivity 1945–46.

If first of all the course of gland infections in *gambiae* is followed through the season, it is seen that there is a very striking inverse ratio between population density and sporozoite rate. The peak production of *gambiae* in June and July is associated with low sporozoite rates, while in October, November and December, when the numbers of *gambiae* are very low, such extraordinary high rates, up to nearly 29 per cent., are obtained that it must be almost entirely a population of old mosquitos. The high infection rates for *gambiae* in Apapa Lemu are very similar to those obtained by Barber in Otta (20 miles inland) where he must have been dealing with a pure *gambiae* population, uncontaminated with *melas*.

If these figures are compared with those for *melas* under the same conditions, the relation between population and gland infection will be seen to be quite different from that of *gambiae*. In fact the chart seems to show that in *melas* the sporozoite rate varies more directly according to the population, an increase in sporozoite rate accompanying an increase in the incidence of females in houses.

Even allowing for the scanty figures in some months, the difference in the course of infection in the two species, in the same village, is sufficiently marked as to suggest some fundamental difference in behaviour. These marked differences in natural infectivity are particularly interesting in the light of Robertson's (1945) observations on the gametocyte threshold for the infection of *gambiae*, in which he found that *A. melas* seemed to be as susceptible to experimental infections as *A. gambiae*.

The relation between sporozoite rate and population density in *gambiae* is almost exactly what would be expected if all the mosquitos were produced beside the village, and spent all their lives within this limited area ; old mosquitos tending to remain in the village long after the breeding grounds have stopped producing adults.

In the case of *melas* one explanation would be that the old females—presumably the most highly infected—tend to remain in the same locality only when suitable breeding conditions are present, conditions associated with a high output of adults. When these breeding grounds become unfavourable, the old females move away from the neighbourhood, and the low population of *melas* in the village is then made up of young mosquitos.

Composition of the Population inside Houses.

Both *melas* and *gambiae* usually take two days to digest the blood meal and develop the ovaries. Thus, if the mosquito feeds, say, on Monday night, it will be ready to lay eggs on Wednesday night. Furthermore, marking experiments show that on the night on which eggs are laid, the female returns for a blood meal the same night. Both species are liable to feed every second night in nature.

If we exclude unfed females (which are mostly newly-emerged and unfertilised) the population inside the houses during the day falls into two distinct groups ; those called "blood-fed" which fed the previous night, and those called "gravid" which fed the night before, and which will be ready to lay eggs that night ; in which respect they closely resemble *A. minimus*, Theo., in Assam (Muirhead Thomson, 1941).

Both in Sierra Leone and Nigeria the blood-feds are always more numerous than the gravids in daytime catches, and sometimes the disparity is very great. In the catching stations round Lagos the two groups were analysed by egg characters, and differences were disclosed between the population of *gambiae* and that of *melas* in the same houses. For example in 16 collections in 6 different stations the following results were obtained :

	Blood-fed	Gravid
melas	128	17
gambiae	257	164

Out of 566 adults identified by egg characters, the proportion of blood-fed to gravid was 7·5 : 1 for *melas*, and 1·5 : 1 for *gambiae*. It is clear that *gambiae* in the gravid state makes use of the house as a resting place to a much greater extent than gravid *melas* ; as far as *melas* is concerned, the consistent wide differences between the number of blood-fed and the number of gravid seems a sure indication that gravid *melas* must make use of resting places outside.

Outside Resting Places.

The knowledge that gravid females at least must have other daytime resting places besides habitations, led to a careful and prolonged search for regular outdoor resting places. With the exception of Blacklock's unusual observation in Freetown (1941), previous search for *Anopheles* resting outside had not been very fruitful in West Africa. Barber, Rice, and Brown (1932) in a thorough search in Liberia found only a single gravid *gambiae* resting out of doors. Search by the author for outside resting places in Freetown Estuary (Wellington village) had produced nothing. In contrast to conditions in West Africa there were the observations of Leeson (1931) in Southern Rhodesia in which almost as many *gambiae* were found out of doors as inside buildings.

A vigorous search was started in villages round Lagos, and after many discouraging results, an increasing number of places used by both *melas* and *gambiae* was finally found (Pl. X, figs. 1, 2, 3).

The results are shown in Tables XI and XII.

TABLE XI.

Anopheles gambiae and *melas* adults in outside resting places.

Locality	Number of positive collections	Type of outside resting place	Blood-fed	Gravid	Unfed	Eggs	
						melas	*gambiae*
Anjorin ...	10	Termite mounds ...	11	13	10	13	1
		Trunk of palm tree ...	1	0	0	—	—
Jinadu ...	4	Termite mounds ...	11	4	6	4	0
Olodi ...	1	Earth well	1	0	0	1	0
Alagbon...	1	Shady bank	0	1	0	1	0
Mekunwen	3	Termite mounds ...	2	1	2	—	—
		Tree hole, mango ...	1	0	0	1	—
		Palm trunk	0	0	2	—	—
Senu ...	3	Buttress roots of forest trees (Pl. x, fig. 3) ...	2	1	20	—	—
Alagbede	1	Trunk of palm ...	0	1	0	—	1
Makoko	50	Shaded earth bank ...	309*	302*			

*(See Table xii.)

From the Table it will be seen that quite a variety of places may be used by blood-fed and gravid females, although not a single female was found on open hedges as observed by Blacklock (1941) in Freetown. Of particular interest are the termite mounds (Anjorin, Jinadu, and New Mekunwen) and the shaded earth banks (Makoko), the only places where anything approaching regular catches could be made.

The termite mounds were under heavy shade of palm thicket (Anjorin, Pl. X, fig. 1) or mango grove (Jinadu). Exposed termite mounds yielded nothing. Mosquitos were found resting in the exposed galleries of mounds which had been broken up by the natives either to get the queen termite, or to get good soil for mud walls. They were also found in deep natural fissures in the mounds ; but the best place was in the large burrows made by ground rodents at the base of the mound. Even in such favoured places it was seldom possible to find more than one or two females per man-hour.

The only place where regular, and occasionally high, catches of mosquitos outside were obtained was the heavily shaded stretch of earth bank beside Makoko village (Pl. X, fig. 2). In this place unfed, newly-emerged, unfertilised females and males were also regularly found, but apart from noting their presence attention was concentrated on the blood-fed and gravid females. In 50 collections extending over 13 months, not only the gravid females to be expected were found, but also an almost equal number of blood-fed females (the actual numbers were 309 blood-fed and 302 gravid) which must either have fed out of doors, or left the house shortly after feeding. Their origin will be discussed in the next section.

In Makoko, too, the outside resting habits of *melas* could be compared with those of *gambiae* under the same conditions, as both species were abundant in the village houses for several months.

The outside catches at Makoko for 6 months in 1945, showing the numbers of *melas* and *gambiae* based on egg determinations are given in Table XII. This Table should be compared with Table II and fig. 1 showing the seasonal incidence of *gambiae* and *melas* in Makoko houses. At the bottom of the Table it is seen that of mosquitos taken in daytime resting places in houses, *gambiae* was twice as abundant as *melas* ; in outside resting places over the same period the position was reversed, five times as many *melas* being taken as *gambiae*.

TABLE XII.

Incidence of blood-fed and gravid *gambiae* and *melas* adults in outside resting places at Makoko village, March–August 1945 (to be compared with Table II showing incidence in houses).

		March	April	May	June	July	August
Blood-fed and gravid		38	36	20	188	151	103
Egg batches laid	*melas*	15	31	13	113	41	30
	gambiae	0	1	2	6	29	5
Total adults	*melas*	38	35	17	179	89	88
	gambiae	0	1	3	9	62	15
Percentage adults	*melas*	100	97	87	95	60	86
	gambiae	0	3	13	5	40	14

Of 536 adults caught outside, March to August 1945, 446, or 83 per cent., were *melas* ; 90, or 17 per cent., were *gambiae*.

In village houses at Makoko, March to August 1945, of 1,772 adults, 32 per cent. were *melas*, and 68 per cent. were *gambiae*.

Proportionately, therefore, it would appear that over the whole period *melas* makes use of outside resting places to 10 times the extent that *gambiae* does. But this may be rather too simple an interpretation of the results, because catches at short intervals show no direct relation between indoor and outside catches. For example it is seen from Table II and fig. 1 that in Makoko houses the highest catches of *gambiae* were recorded in May and June, and yet it was not till July that *gambiae* was taken outside regularly.

Table II shows similar discrepancies for *melas* in the month of June 1945, when the proportion of *gambiae* to *melas* in houses changed greatly after the middle of the month, the number of *melas* increasing roughly 10 times ; the mean daily catch outside showed only a slight increase.

These discrepancies raise many interesting questions. Does the proportion of *melas* to *gambiae* in houses represent the real proportion in the whole existing population, or does the outside catch give a better sample ? In many ways Makoko is exceptional in having ideal outside resting places just beside an isolated village. In most places the outside resting must be exceedingly scattered, and the recording of a single mosquito in thick bush, for example, assumes great significance. There are large villages, such as Apapa Lemu, where suitable outside resting places are scarce, and where not a single mosquito has been found outside despite regular high catches in the houses. In such a village the proportion of blood-fed and gravid females in houses remains much the same as in a village where ideal outside resting places exist (Table XIII), suggesting that mosquitos are still leaving the houses for outside resting places which have not yet been detected.

TABLE XIII.

Composition of *Anopheles* population in houses (daytime catch) in relation to proximity of suitable outside resting places.

Village	Remarks	Total catch	Per cent. blood-fed
Makoko ...	Ideal outside resting places beside village	230	82
Apapa Lemu ...	Ideal outside resting places absent ...	120	85

From a practical point of view these outside resting places are of much more than academic interest. Sporozoite infections in both *gambiae* and *melas* caught outside have been recorded (Table IX); and the outside catches are also of the greatest importance in interpreting the results of malaria control by house spraying with DDT (Muirhead Thomson, 1947).

Movements of Mosquitos in Houses.

Previous workers have observed the considerable daily turnover in the *gambiae* population in houses (Gordon & others, 1932, in Sierra Leone; Barber & others, 1932, in Liberia; Causey & others, 1943, in Brazil). In the present study it is hoped to carry the enquiry further, firstly, by paying more attention to the behaviour in relation to different stages in the gonotrophic cycle, i.e. whether the female is unfed, blood-fed, half gravid, or gravid; and secondly, to observe the natural movements of mosquitos in and out of houses, and to avoid basing conclusions on the possibly unnatural movements of stained and liberated mosquitos.

In the section on composition of the population in houses it was deduced, from collections alone, that many gravid females must make use of outdoor day-time resting places. Catching in outside resting places showed that not only were gravid females found, but also an equal number of blood-fed mosquitos, which may have fed inside or outside. From these findings alone it appears that there may be two or three different times during the 48-hour gonotrophic cycle when the female is liable to leave the house.

If houses are searched in the early hours of the evening, after sundown, only a few females can be found in rooms where regular high day-time catches are being recorded. Observations in the laboratory show that mosquitos, both laboratory bred and wild caught, show great activity at dusk, and that even those females which are not ready to lay eggs are strongly attracted towards the waning light coming in from outside. This is not peculiar to *melas* and *gambiae*, but has also been observed in other Anophelines, such as *A. minimus* (Muirhead Thomson 1941) in Assam, and in *A. culicifacies*, Giles (Rajindar Pal, 1945).

This strong attraction towards the outside light at dusk was used as the basis of a simple but most effective way of trapping mosquitos leaving a house at that time, and was later extended for use at any time throughout the night and at dawn.

Several huts were constructed with mud walls and palm thatch roof. (The actual dimensions were, 12 ft. long, 6 ft. broad, 4 ft. high at eaves, 6 ft. to top of roof.) Two boys slept regularly in each hut. Apart from slight modifications, described below, this type of mud and thatch hut was typical of African village houses in this part of the coast. The palm thatch was sufficiently thick to prevent any light from shining through, and at the same time there was easy access to hungry mosquitos through small fissures and chinks in the eaves. The inside of the door was covered with a dark curtain. Over the inside of the single small window opening, black cloth was fitted in which an opening of 1 ft. square was cut. Over this opening a trap cage of 1 ft. cube, made of light wire framework and white mosquito netting, could be fixed from outside (Pl. X, figs. 4 and 5). One side of the trap cage was inverted to form a funnel narrowing to a small opening, about $\frac{1}{4}$ inch diameter, inside the cage near its front wall. The narrow end of the funnel was suspended inside the cage by stout cotton threads running to the four front corners. The cage was provided with one or two sleeves at the sides for removing mosquitos and it was arranged in such a way that from inside the hut, at any time between dusk and dawn, the only light coming in from outside was through the 1 ft. square window opening. Observations at dusk showed that mosquitos wanting to leave the hut were strongly attracted to the window trap, and quickly found their way into the cage from which there was no escape.

From the strength of the attraction to the faint light it seemed almost certain that the mosquitos were not leaving the house by any other way ; this was confirmed by hanging a very large muslin net over the whole hut, and lowering it before dawn, in which case 50 or 60 females could be taken in the window trap, but not a single one inside the large net.

Dusk observations.—Before describing these findings it would be as well to recall the convention adopted here of naming the different stages in the 48-hour gonotrophic cycle of *melas* and *gambiae*. Briefly, a female feeding on Monday night will be called blood-fed when caught on Tuesday. Captured at night any time between dusk on Tuesday and dawn on Wednesday it is called " half-gravid " ; caught during the day on Wednesday it is called " gravid ".

In these dusk observations an experimental hut was used in which the mosquito population had been left undisturbed during the day. About an hour before dusk the doors were closed and the window traps fixed in position over the window openings. About an hour after sundown counts were made of the half-gravid females (those which had fed the previous night) caught in the window cage, and of the residue still resting in the hut. The window cage also contained many gravid females leaving the hut to oviposit ; these were naturally excluded from the count. Inside the hut the population was almost entirely half-gravids, it being too early for blood-fed mosquitos to be found, and all gravid females having left the hut.

The results of 7 experiments, involving *melas* alone, showed that of 277 adults, 197 were taken in the window trap, and 80 in the hut.

It is seen that under the ideal conditions offered to mosquitos resting in the hut, approximately 70 per cent. of the half-gravid *melas* leave the hut at dusk. It is fairly certain that in the usual African village house, where mosquitos are much more liable to be disturbed, and where open doors and wide eaves present obvious means of exit, the proportion of half-gravid females remaining in the hut will be even smaller.

Dawn observations.—In the outside collections not only were the gravid females to be expected found, but also an equal number of blood-fed females, which may have been mosquitos that had fed out of doors, or those which fed inside houses and left before daytime. To test the latter possibility window traps were fixed in position about two or three hours after dark. About two hours after dawn next day the catches in the cage and in the hut were compared. It would be as well to point out at this stage that in these huts there was no indication of blood-fed or half-gravid females entering the huts from outside. Empty huts with doors closed and window traps in position yielded nothing, at a time when high catches were taken in adjacent occupied huts. It was certain, therefore, that this method gave an accurate indication of the proportion of blood-fed females likely to leave an ideal dark hut after feeding.

The results of 8 experiments involving 224 *melas* and 23 *gambiae* show that the proportion varies from day to day, but in this series as a whole 43 per cent. of the *melas* which fed in the hut left at dawn, while for the small number of *gambiae* the figure was 26 per cent.

Again it must be stressed that conditions for daytime resting in the hut were ideal, with the minimum of disturbance. In the ordinary native hut the activity of the inhabitants at dawn would certainly result in an even higher proportion of the freshly blood-fed females leaving the house.

Even though *gambiae* and *melas* are domestic mosquitos, in that they may be found resting in numbers inside houses by daytime, it is clear that the day catch may represent only a small fraction of the total number feeding there at night.

The partial exodus at dawn is of the greatest practical importance in house spraying with insecticides, as it can be shown that with regular Pyrethrum spraying the proportion of blood-fed females leaving the house is greatly increased, while for a house treated with DDT, all females may leave after feeding (Muirhead Thomson, 1947).

Somewhat similar behaviour was shown by several other Anophelines, *funestus*, Giles, *pharoensis*, Theo., *coustani*, Lav., *hargreavesi*, Evans, *paludis*, Theo., and *obscurus*, Grünb. Of 50 blood-fed *funestus* females caught in 95 collections, 9, or 18 per cent., were caught in the window trap. For *coustani* the figure was 71 per cent.

As will be shown in a later section, the proportion of blood-fed females leaving the hut at dawn may also be influenced by the nature of the host, animal or man.

Feeding Activity at Night.

A preliminary series of catches in native village houses at various times between dusk and midnight showed a great scarcity of *melas* and *gambiae* in rooms where regular high day-time catches were being made. In these evening catches comment has already been made on the small number of half-gravid females remaining in the houses, due to so many leaving at dusk. During this period there is also a remarkable scarcity of freshly blood-fed females, indicating very little feeding activity in this early third of the night.

To study this question in detail the huts described in the earlier section were used, and collections were made, mainly by the author and one collector, at 2-hourly intervals from 8 p.m. till dawn. The same two boys slept in the hut each night.

The experiment is roughly on the same lines as that of Haddow (1942) on *gambiae* and other mosquitos in East Africa, but there are differences in detail. In this series of collections blood-fed females were concentrated upon, giving results which show time of feeding rather than time of entry. As far as *melas* and *gambiae* were concerned, unfed females could not be differentiated, as they require a blood meal before they can lay identifiable eggs.

The collections made at dawn included not only blood-fed females resting in the hut, but also those caught in the window trap as they leave the hut. Window trap inspections through the night showed little indication of freshly fed females leaving the hut during the hours of complete darkness.

TABLE XIV.

Blood feeding activity of *melas* and *gambiae* in human habitation at night. Results of 9 all-night catches in hut. June and July 1945.

	Period of night				
	8–10	10–12	12–2	2–4	4–dawn
Blood-fed adults *melas*	13	27	100	97	154
Blood-fed adults *gambiae*	2	3	20	16	28
Percentage of total night catch. Both species	3	7	26	25	39

The results of 9 all-night collections made in June and July 1945 are given in Table XIV.

They show that for *melas* and *gambiae* together, 90 per cent. of the blood feeding takes place after midnight, the period of greatest activity being between 4 a.m. and dawn. The figures for *gambiae* are rather small, but they show the same general trend as *melas*, and at the same time show differences from behaviour in East Africa as recorded by Haddow (1942). Haddow found that the number of fed females caught up to midnight was 33 per cent. of the total night catch of fed females. In the present experiment only 7 per cent. of the feeding took place before midnight. In this respect it should be pointed out that in catches in the early evening some of the half-gravid females found, i.e. those which fed the previous night, still contain a lot of blood, and may easily be confused with freshly fed females unless closely examined. Kerr (1933) in West Africa found that "*gambiae*" (almost certainly mixed *gambiae* and *melas*) were most active from midnight onwards.

In these experiments it was hoped to get information about the feeding activities, not only of *gambiae* and *melas* in general, but also more precise data about infected mosquitos. Unfortunately in this locality, the only one suitable for such an experiment, the sporozoite rate of *melas* was only 0·3 per cent. (Table IX), and therefore dissections of mosquitos caught at different times of the night yielded nothing of interest.

In these night collections the apparent scarcity of hungry unfed females was striking—in 9 collections only 25 unfed as compared with 462 blood-fed. The hungry females may possibly be more active than the gorged ones, or they may be more readily disturbed by the beam of the torchlight. It was not possible to determine the extent to which *melas* and *gambiae* settle on the inner walls or ceiling of the room before biting, but later experiments on house spraying with DDT suggested that most mosquitos fed without first settling on the walls.

In these night collections at Lagos the increasing tempo of biting between midnight and dawn was very consistent irrespective of the type of night, such as bright moonlight or complete darkness. The moonlight certainly had no obvious effect on feeding activity. As for the rate of entry, which may be quite a different thing from feeding activity, Ribbands (1946) has shown that *funestus* at least may be strongly influenced by moonlight.

When describing above the hut used in this experiment, it was shown that with the door closed and the window trap in position, there was no indication of blood-fed or gravid females entering the hut from outside at dawn or in the early morning. In the huts used by Haddow (1942) there was an open space 1 ft. high round the top of the wall, and this difference in hut construction probably accounts for the large number of mosquitos which entered his huts at dawn seeking shelter for the day.

Host Selection by *A. melas* and *A. gambiae.*

In order to test the feeding activities of *Anopheles* on animals other than man, improved designs of baited mosquito traps have been suggested from time to time. Bates' (1939) modification of the Magoon type of trap seems to offer distinct possibilities, particularly with " wild " mosquitos which are apt to leave the shelter in which they feed.

In the present investigation, dealing with species in which a varying proportion of the females may leave the house or hut after feeding, the experimental huts described earlier in this paper provide a very efficient trap. With its detachable window trap cage, the hut traps all the mosquitos whether they remain in the hut after feeding, or whether they attempt to leave for outside. It has already been pointed out that this hut with its door closed and covered with a curtain inside, and with the window trap in position, offers no obstacles to hungry mosquitos entering through narrow chinks in the eaves ; but at the same time there is no indication of any blood-fed mosquitos entering the hut from outside in search of a day-time resting place. It is, therefore, certain that all the blood-fed females found in such a hut have actually fed there, whereas in the ordinary open type of man or animal habitation the origin of the blood-fed females found is uncertain.

An attempt will be made to show in the course of the following experiments that this type of hut trap has many advantages over the Bates trap referred to above in studying the feeding reactions of all *Anopheles*, whether wild or domestic, and also in that it reveals that the subsequent behaviour of the mosquito may be affected by the type of animal on which it has fed.

In all these experiments from 2 to 6 identical huts with window traps, exactly as described above, were used.

In the first series of experiments mosquitos were offered the choice of feeding on 2 men—always sleeping in the same hut—and an increasing number of animals.

Experiment 1. Two identical huts used. One occupied by 2 men, the other by 4–6 goats.

Experiment 2. Three huts used. One occupied by 2 men. One by 4–6 goats, the third by 4 pigs.

Experiment 3. Four huts used. One occupied by 2 men. One with goats as above. One with pigs as above. One with a cow.

The results of these are shown in Table XV.

TABLE XV.

Blood-fed *melas* and *gambiae* caught in man and animal baited hut-traps. Mosquitos offered choice of feeding on fixed number of men (2) and increasing number of animals.

Number of collections	Man (2)		Goat (4–6)		Pig (4)		Cow (1)	
	Hut	Window trap	Hut	Window trap	Hut	Window trap	Hut	Window trap
12	25	11	3	55				
26	349	117	10	183	89	58		
19	100	26	5	31	28	15	117	9
Total ...	474	154	18	269	117	73	117	9
Eggs laid *melas*	205	64	1	96	52	28	42	6
Eggs laid *gambiae*	11	1	1	12	0	0	0	0

Considering the totals first, i.e. hut catch and window trap catch added together, it is seen from Experiment 1 that 4 goats were rather more attractive to *melas* at least than 2 men. From Experiment 2 it is seen that despite the increase in the number of animals, the 2 men attracted 58 per cent. of all mosquitos. In Experiment 3 it is seen that the addition of a large cow to the collection swayed the balance the other way, and only 38 per cent. of the females fed on man.

These results apply mainly to *melas*, but the small numbers of *gambiae* recorded were found only in the man hut and goat hut. With *melas* it appears that with the choice available it may feed indiscriminately on man, goat, pig, and cow. It is evident that with such an excess of animals there may be considerable deviation of the mosquitos from man, but at the same time the proximity of so many animals is not sufficient to prevent *melas* feeding regularly on man.

If the hut catch is now considered as distinct from the window trap catch, one striking fact appears. With man, pig, and cow the proportion of blood-fed *Anopheles* leaving the hut at dawn varies from about 10 per cent. to about 40 per cent. In the goat hut, however, 94 per cent. of 287 blood-fed *Anopheles* were taken in the window trap as they endeavoured to leave the hut, and on most days not a single blood-fed female was found resting inside the hut. Although the huts were identical there was always the possibility of the mosquitos being influenced by some unknown factor. To test this the goats were removed from the hut, and a fortnight later 2 boys started sleeping in the hut at night. Of 89 *melas* taken in 5 collections, the proportion taken in the window trap was now 27 per cent. It is clear, therefore, that while the goats are suitable hosts for *melas*, *gambiae*, and *funestus*, the presence of these animals renders an otherwise suitable day-time shelter unattractive to the mosquitos which have fed in the hut, and nearly all of them leave the hut after feeding. Going on the hut catch alone, without window trap, one might have reached quite erroneous conclusions about the suitability of this animal as host.

In a second series of experiments the numbers of animals were kept fairly constant (3–4 goats, 4 pigs, and 1 cow or calf) while the number of men was increased from 2 to 4, and finally to 6, with 2 men in each hut Again the results apply mainly to *melas*, which formed 95 per cent. of the *melas-gambiae* group.

The results obtained, together with those of Experiment 3 above, are combined in Table XVI.

TABLE XVI.

Blood-fed *melas* and *gambiae* caught in man and animal baited hut-traps. Number of animals constant. Number of men increasing from 2–4–6 (95 per cent. of adults were *melas*).

Experiment	Number of collections	Total feeding on man	Total feeding on animals	Percentage feeding on man
2 men (1 hut) and 8 animals (3 huts)	19	126	205	38
4 men (2 huts) and 8 animals (3 huts)	5	236	27	90
6 men (3 huts) and 8 animals (3 huts)	5	702	11	98

They show that when the number of men increases there is a sharp decrease in the numbers of *melas* feeding on animals, till finally when 6 men are available in 3 huts, the numerous animals have almost ceased to attract any *melas*, and the deviation by animals fades out.

Needless to say one could plan an almost endless variety of experiments on these lines. For example it would be interesting to see if the 6 men were so much more attractive because they were spaced out in three huts. One would also like to augment these observations by precipitin tests on *Anopheles* found in outside resting places.

In the course of these experiments some information was also gained about the feeding habits of other species of *Anopheles.*

A. funestus.—Forty-four blood-fed *funestus* were taken in the course of Experiments 1–3, 18 from man hut and 26 from goat hut. Of those feeding on goat all but 3 were taken in the window trap, reacting to this animal in the same way as *melas.*

A. gambiae.—In Experiments 1–3 egg identifications showed that about 67 *gambiae* were found, 28 in the man huts, and 39 in the goat hut. This species agrees with *funestus* in that not a single one was found feeding on pig or cow in a large number of collections.

A. coustani.—In Experiment 3 the number of blood-fed *coustani* taken in the man, goat, pig, and cow huts was 22, 2, 10, and 185, respectively, showing a marked preference for cow as a host. Of those feeding in the huts, the number leaving after feeding was roughly the same as that remaining.

Of the various animals used in the experiments above, goat is the one most constantly present in the villages round Lagos, and the one most closely associated with man as it spends most of the night sleeping under the eaves of the village houses.

Pigs are abundant in villages in the 5-Cowrie Creek area, but are not conspicuous elsewhere. Cattle are comparatively scarce except in odd places where herds may be kept in open paddocks at night. There are no real animal houses as such, the most that is provided is a rough thatch shelter with no walls. In this part of the coast, therefore, animals are seldom present near habitations in such an excess as occurred in these experiments, although there are other parts of Nigeria where a greater number of animals may make deviation more effective.

The experiments described above indicate that " host preference " can be a very fluid term, the preference of any mosquito for man or animals being liable to vary enormously depending on the proportion of the hosts available. A series of precipitin tests on blood meals of mosquitos caught in outside resting places would be a most valuable contribution, although in the Lagos area the same difficulties would be encountered, as when dealing with the races of *A. maculipennis*, that with a mixed population of *gambiae* and *melas* it is not possible to identify the blood meal and the mosquito at the same time.

Mating Habits.

Observations in nature.—Swarming of " *gambiae* " outside was first observed at Makoko village, 5 miles from Lagos, in June 1945. The composition of the mosquito population in nearby houses at this time was 90 per cent. *gambiae* and 10 per cent. *melas*, while *melas* was the dominant mosquito in outside resting places. As no females were caught in these swarms it was impossible to tell to which species they belonged, and they are therefore referred to as " *gambiae*."

The first swarm was seen at dusk on 6th June, about 7 feet above the ground, over a small palm shrub. Males went on swarming till it was too dark to see them.

On 9th June swarming was again observed at exactly the same place. About 30 males were caught in three or four sweeps of the net. No swarming was seen anywhere else in the vicinity, although there was an enormous production of *gambiae* from a large borrow pit a few yards away (up to 19 pupae per dip).

On 12th June swarming was seen at dusk at exactly the same place, and three other swarms were seen over small palm bushes or shrubs a few yards away.

On 17th June a single swarm was seen over a different bush from that of the 9th. One sweep of the net ended the swarm completely. No females were found. Swarming was seen for the last time on 24th June.

Observations in the laboratory.—Attempts to induce *gambiae* and *melas* to mate in captivity had met with no success in Sierra Leone. All sorts of cages had been used indoors and outside at dusk, and using large numbers of laboratory bred adults. The females were fed regularly on the author's arm, but not a single egg was laid, and it is evident that with both these species repeated blood meals fail to develop the ovaries to maturity in unfertilised females. These consistently negative results in West Africa are even more interesting in view of Macgregor's (1923) observations in Mauritius in which he had no difficulty at all in getting *gambiae* and other Anophelines to mate readily in captivity.

In Lagos the same procedure was repeated with no success, till finally the cages full of mosquitos were kept in a living room after dark, and illuminated by a table lamp with an orange-coloured shade. In these 1 ft. cube cages swarming, mating, and fertilisation now took place, and when the females were given blood meals oviposition followed in due course.

All *gambiae* adults bred out in the laboratory from *gambiae* type eggs, laid *gambiae* type eggs in turn. Similarly *melas* produced eggs true to form, giving additional proof of the reliable nature of this diagnostic feature. With all these fertilised females a single blood meal was sufficient to develop the ovaries and bring about oviposition.

The history of one of the laboratory reared and fertilised *gambiae* females is particularly interesting. It lived for 12 weeks (86 days) in a small 6×6×6 inch cage. During that time it took 30 blood meals and laid 28 batches of eggs. All the batches were distinct *gambiae* type eggs.

Cross fertilisation experiments.—As *gambiae* and *melas* could now be induced to mate in captivity, it opened up the possibility of experiments in cross-fertilisation in order to establish the exact relation between the two species.

Using the same methods as above, pure strains of *melas* and *gambiae* were bred out in the laboratory, and as soon as the adults emerged *gambiae* males and *melas* females were put in one cage, and vice versa. The number of successful cross matings was small, 7 *melas* females being fertilised by *gambiae* males, and 2 *gambiae* females being fertilised by *melas* males. All these females laid fresh batches of eggs after each blood meal.

The form of egg laid is evidently determined by the female parent, and there were no transitional forms. The eggs from both crossings produced healthy larvae—whose reactions to salinity are described later in this section—and eventually a generation of healthy hybrid adults.

Attempts to induce the hybrid adults to mate with each other were unsuccessful, although by itself this negative finding is not proof of mutual sterility. Examination of 31 males from the *gambiae* male-*melas* female crossing showed testes with varying degrees of underdevelopment. None of them had the coiled masses of filiform sperms such as are found in normal *gambiae* and *melas*. In many the testes were actually atrophied, and in only a single case were recognisable sperms seen. *Melas* females were exposed to these hybrid males; after a week one female laid a batch of *melas*-form eggs; after a further blood meal another batch was laid, but none of the 50 eggs in either batch hatched.

Examination of 4 males resulting from the *melas* male-*gambiae* female crossing showed a further degree of underdevelopment in the testes, which were reduced to a shred of tissue.

In the female hybrids from both crossings the ovaries presented normal appearance, but attempts to cross them with pure *gambiae* males were unsuccessful, even though the ovaries develop well towards maturity with frequent blood meals.

The results are shown in diagrammatic form in fig. 3.

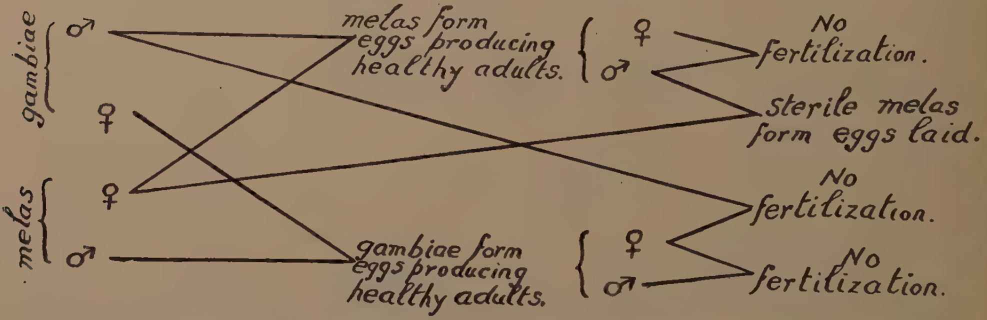
gambiae
♂
♀
melas
♀
♂
melas form eggs producing healthy adults.
♀
♂
No fertilization.
sterile melas form eggs laid.
No fertilization.
gambiae form eggs producing healthy adults.
♀
♂
No fertilization.

Fig. 3.—Diagram showing results of cross-fertilisation experiments with *A. gambiae* and *A. melas*.

As there seemed every indication that the hybrid males were sterile while the females were normal, the relationship between *melas* and *gambiae* resembles that between *atroparvus* and *labranchiae*, or between *atroparvus* and *subalpinus*, in the *maculipennis* group (Hackett, 1937, Bates, 1939).

To return to the hybrid larvae, these were tested for salinity reactions. Ribbands (1944) has shown that the reactions of *melas* and *gambiae* larvae to sudden changes in salinity can be used as a test for the species. This test was applied to larvae produced from the *gambiae* male-*melas* female crossing, the *melas* male-*gambiae* female crossing, and pure cultures of *melas* and *gambiae* larvae as controls. Larvae were put in 37½ per cent. sea water (11·9 gm. NaCl per litre) for 24 hours, then transferred to 75 per cent. sea water (23·8 gm. NaCl per litre) for 24 hours.

The results are shown in Table XVII, in which it is seen that hybrid larvae from both crossings, whether laid by *gambiae* females or *melas* females, were fairly tolerant of salinity changes which killed all pure *gambiae* larvae. In fact the hybrids appear to be as tolerant as pure *melas* in this respect.

TABLE XVII.

Reactions of *melas*, *gambiae*, and hybrid larvae to changes in salinity. Ribbands' test—larvae put in 37½ per cent. sea water for 24 hours, then transferred to 75 per cent. sea water for 24 hours.

Larvae		Number tested	Number surviving
melas		20	16
gambiae		20	0
Hybrids	From *melas* female crossed *gambiae* male ...	10	7
	From *gambiae* female crossed *melas* male ...	10	8

Breeding-places of *A. melas* in the Lagos Area.

In a study of the breeding habits of *melas* in Freetown Estuary, Sierra Leone, the author found a striking association between *melas* breeding and *Avicennia* mangrove (Muirhead Thomson, 1945). With the exception of one or two small patches of *Paspalum*, dense breeding of *melas* was mainly confined to comparatively few *Avicennia* "orchards", while the great bulk of the mangrove (*Rhizophora*) and its adjacent *Paspalum* areas were almost entirely harmless. There real estuarine conditions were found with large rivers pouring in great quantities of fresh water, and a great deposition of silt taking place where the river water and the sea water met.

In Lagos, on the coast of Nigeria, rather different conditions are encountered. Interminable lagoons stretch along the low-lying coast, and except for narrow outlets are separated from the sea by comparatively narrow sand spits. There are few large rivers opening into the lagoons near Lagos, and true estuarine conditions with maximum growth of mangroves are absent.

One characteristic feature of the tidal swamps round Lagos are great stretches of *Paspalum* grass which have been well-known breeding places of the "melanic coastal variety" for some time (Barber, 1931). An extensive scheme of tidal swamp reclamation by "bunding" was started in 1943 (Gilroy & Chwatt, 1945), but fortunately many of these swamps had been seen in their natural condition, before reclamation, and the conditions observed under which *melas* was breeding.

For a long time there has been considerable felling of mangroves round Lagos and it seemed possible that some of the dangerous *Paspalum* swamps were originally covered with *Avicennia*. This is certainly true in some cases, where the *Avicennia* trunks are still visible. In other places much of the *Paspalum* is associated with *Rhizophora*, but there are still great stretches of *Paspalum* in which there is no sign of any original mangrove formation.

As for *Avicennia* orchards, there are, or rather there were until very recently, some very well marked ones particularly at N.E. Ikoyi and on Iddo Island, that closely resemble the orchards in Freetown Estuary. The rest of the tidal area is made up of *Rhizophora*, small in most places, and never reaching the profusion found in the Freetown creeks.

A thorough search for breeding-places was carried out in the first three months of 1945 at a time when *melas* was dominant in most of the Lagos area. The great bulk of *Paspalum* and mangrove yielded practically nothing, but here and there real breeding grounds were found where, over a limited area, larvae were very abundant. Such breeding grounds occurred in the following places :—

Inside *Avicennia* orchards (N.E. Ikoyi, Iddo).

In *Avicennia* and adjacent *Paspalum* (Iganmu, Makoko, Apapa Lemu).

In unusually large crab holes among *Paspalum* (Okesuna).

In *Paspalum* alone (Anjorin).

Relict salt pools at swamp edge (Mekunwen).

Paspalum in open spaces among *Rhizophora* (Anjorin).

Salt ponds (Alagbon).

From this it is seen that in the Lagos area there is no sure guide to important breeding grounds, except that unbroken *Rhizophora* with no *Paspalum* is consistently negative. *Avicennia* ceases to be an indicator, and the only way to track down the breeding places is to look everywhere. From the point of view of routine control this would be an almost impossibly arduous task ; fortunately the comprehensive nature of the tidal swamp reclamation should eventually eliminate all potential breeding places of *melas*.

When considering the seasonal fluctuations in *melas* breeding as influenced by tides and rainfall, very different conditions again are found from those of Freetown Estuary. In Freetown the tides are sufficiently regular to enable their heights to be predicted months ahead. By studying the tide tables it was possible to forecast the exact day and hour on which the greatest density of *melas* breeding would be encountered in a particular *Avicennia* orchard. In Lagos, on the other hand, the movements of the tides were irregular, and their heights unpredictable. Never at any time was it possible to tell whether the breeding grounds would be flooded or not at the next spring tide. Furthermore, there were distinct tidal variations between parts of the lagoon ; on the same day one breeding ground would be flooded, while another only a few miles away would be untouched. It is this irregular and unpredictable nature of the spring tides which accounts for erratic incidence of *melas* in houses throughout the year, and also for the wide differences between different catching stations in the same month (fig. 1).

How far the spring tides are influenced by rainfall or by unusually heavy showers it is difficult to say, but in some catching stations such as Alagbon and Apapa Lemu, the fluctuations in seasonal incidence do appear to bear some relation to rainfall.

Breeding-places of *A. gambiae* in the Lagos Area.

The various types of breeding-place used by *gambiae* in and around Lagos have been described by Barber and Olinger (1931). Many of these observations have been repeated and confirmed in the course of the present work, in particular the absence of *gambiae* from many different types of water, such as extensive wooded swamps, permanent lily ponds, and also from many, but not all, large grass-edged brick-field borrow pits.

There are one or two *gambiae* breeding-places which are of particular interest.

Wells.—In the dry weather many villages outside the urban area of Lagos, Yaba, and Apapa, have to rely on wells for a water supply. They are particularly noticeable in Idioro just beyond Yaba, and at Olodi village little over half a mile from the Apapa European Reservation. In most of these open earth wells the water surface is 6–10 ft. below ground level, and when they contain deep water they are either completely negative, or only yield a very few young larvae. But occasionally one of these wells, no longer in use, may dry up slowly till a small puddle is left at the bottom. In such a pool 40 or 50 full-grown *gambiae* larvae as well as pupae may be found. In Olodi village only 1 well in 25 could be regarded as dangerous, but there are a great number of *gambiae* adults found in houses in the driest months of the year. There were no other available breeding-places near this arid village. Only *gambiae* eggs have been recovered from these wells.

Although the majority of these wells are harmless, as Barber and Olinger found, the possibility of the odd disused well producing large numbers of *gambiae* in the dry weather may be important in villages situated in arid regions away from the coastal fringe. In examining them it was necessary to use a light ladder, and after descending allow a few minutes for any submerged larvae to come to the surface.

Sedge and other fresh water swamps.—Most of the fresh water swamps on the mainland near Lagos are well wooded and mainly harmless. But there is one extensive exposed fresh-water swamp west of Iganmu, on the Ebute Metta-Apapa Road. The lower part of the swamp is nearly always flooded with deep water, has abundant water lilies, and is not important. But the upper, and more inaccessible end of the swamp, near the villages of Alagbede and Senu, is not permanently flooded, and heavy showers towards the end of the dry weather may produce innumerable shallow puddles on mud over a wide area. In favourable conditions, such as occurred in the early part of 1945, this may become the most extensive breeding ground of *gambiae* in the whole Lagos area. When breeding is at a peak here, conditions are very like those found in the inland swamps of Sierra Leone when the dry weather has reduced the water to shallow surface pools on thick soft peaty ooze (Muirhead Thomson, 1945). The larvae found in these pools may be deeply pigmented, resembling those of *melas*, but all the eggs collected are those of *gambiae*. From the extent of the breeding area, and from the high *gambiae* population in the villages of Alagbede and Senu, it is possible that this swamp was the origin of the *Anopheles*—mainly *gambiae*—invading Railway Compound in March and April 1945.

A breeding ground of somewhat similar type, but on a very reduced scale, is a small patch of sedge swamp fringing the upper edge of the mangroves at Apapa Lemu. Over an area of about 20 or 30 square yards dense breeding of *gambiae* occurred for many months on the thin film of water overlying mud so soft and deep as to be almost inaccessible. This small breeding ground, which accounted for the regular high catches of *gambiae* adults in Apapa Lemu, is within reach of the highest spring tides, and in fact starts just where the *Rhizophora* ends. Egg collections show that it is entirely *A. gambiae* here.

The effect of tides on this breeding-place is particularly interesting with reference to the fluctuations in population of *gambiae* adults in the village (Table III). During the period May to September, which includes the main rainy season, and also the short dry season of August, none of the spring tides was high enough to flood the ground, and breeding went on unchecked. Towards the end of September there were very high spring tides which completely flooded the sedge patch, and brought the dense breeding to an abrupt end. The catches of *gambiae* adults in October were very low, and remained low in November and December while further high spring tides flooded the breeding ground at intervals. The output of *gambiae* in these three months must have been exceedingly low, as the few adults found in the village houses had such high sporozoite rates that the population must have been made up almost entirely of old females.

From about the end of December the spring tides were not high enough to reach the breeding ground, and in the numerous pools now left undisturbed *gambiae* breeding again became profuse, resulting in the high house catches of January 1946. Although well into the dry weather, sufficient water had been left by the previous high spring tides to maintain breeding, in the absence of rainfall, till the end of January. After which time, in the absence of further high spring tides, the breeding grounds dried up, and the house catch of *gambiae* fell off sharply in February and March. By April, drainage and reclamation finally eliminated this patch of swamp.

The local influence of tides on *gambiae* breeding, as distinct from *melas*, has a counterpart in the Scarcies area of Sierra Leone (Muirhead Thomson, 1945) where breeding-places for *gambiae* were produced and maintained right through the dry weather by periodic flooding of the rice fields when high spring tides caused the river to flood over its banks.

Rain-filled pools and puddles.—In a flat low-lying land like the Lagos lagoon area, subject to a heavy rainy season, innumerable rain-filled pools might be expected to persist for months over the whole area, and provide countless breeding-places for *gambiae.* It is true that immediately after heavy rain Lagos island appears as if it can hardly keep its head above water. Fortunately, due to the porous sandy soil, the water drains away very quickly, and it is only in certain districts that rain-filled pools and puddles persist to any extent.

A survey of such breeding-places was made in June and July—a period of heavy rain—in the urban areas of Lagos Island, Ebute Metta, Iddo Island, and Apapa. It is in such temporary pools that the densest breeding of *gambiae* is found, and it is this type of breeding place that is particularly common in streets and compounds.

The greatest number were found in Ebute Metta West. A good many were found in Ebute Metta South, with the numbers decreasing rapidly towards Yaba. During the rainy season the same pools tend to produce large crops of *gambiae* repeatedly, judging by the number of pupae found in these pools. The number of dangerous pools is comparatively small, for example about 50 or 60 in Ebute Metta West, the worst suburb. In June and July there are times when over 300 pupae can be taken in one collection from a small puddle a few feet square, and for several successive days the same pool may yield over 100 pupae per day. These short periods of intense *gambiae* output alternate with periods when the pools are either dry or washed out after heavy rain.

The worst puddles are not in compounds as might be expected, but alongside main roads, and in flooded wheel ruts in the unmetalled side roads in the suburb. There is evidence that some of the worst puddles may appear in exactly the same place year after year. Where roads have been made up, and lateral drains installed, this type of breeding-place tends to disappear automatically. There are parts of Ebute Metta where, in the short course of this investigation, progressive urbanisation has eliminated the potential breeding grounds from whole streets, and as this process continues, the rainfilled breeding-places must automatically disappear.

South of Ebute Metta suburbs, in the direction of Apapa, the sandy soil and the extensive sand filling that has gone on for many years, have combined to make this kind of breeding-place very uncommon. In the extensive dock area of Apapa only one or two puddles of this kind have been found at the height of the rainy season. The high catches of *gambiae* in Apapa Lemu during the rainy season were not due to temporary rain-filled puddles, but to the small patch of sedge swamp described above.

Iddo Island, too, has remarkably few of these pools which are so common in Ebute Metta a short distance away, and a remarkable scarcity of *gambiae* breeding-places of any kind.

Lagos Island is particularly interesting. Reference has already been made to the high catches of adult *gambiae* in houses near N.W. Marina and to the scarcity in the central part of the town. In the present investigation careful search during the rainy season showed a great scarcity of *gambiae* breeding-places over most of the island west of the canal. The few persistent pools were mostly negative, and great areas remained completely free of pools right through the rainy season.

Barber and Olinger (1931) found *Anopheles* larvae and pupae in every one of 5 surveys in both wet and dry seasons. But the number of breeding-places was so small that they did not think they could account for all the adults found in houses.

In the N.W. Marina area only one major breeding-place has been found, but it is of such a unique character that not only does it easily account for all the adults found in the neighbourhood, but it should also stand for all time as an object lesson to malariologists in West Africa. This breeding ground took the form of a vegetable garden 45 ft. square. The muddy soil was raked into mounds for cultivation. Between and around the mounds was a series of shallow pools swarming with larvae of all stages, and pupae, in densities never before encountered. The compound was first discovered at the end of July, and in about 1½ hours 3 collectors took 1,320 pupae from the pools, and the pools were still swarming with pupae when they left. A few days later, using a wooden frame 1 ft. square, pupae were collected from three parts of the compound. In one sample 730 pupae were taken, and the mean was 380 per sq. ft. In all samples there were still many pupae left in the liquid mud. The area of breeding ground was approximately 300 sq. ft. As pupal mortality is negligible, this figure can be used to give an estimate of the *gambiae* output from the compound. Assuming half the adults to be males, the remarkable figure is obtained of an output of about 50,000 female *gambiae* per day from this one compound, a state of affairs which persisted for several weeks.

Collections from these pools yielded eggs of *A. gambiae* in 129 cases and of *A. melas* in 3.

The occasional occurrence of very small numbers of *melas* eggs in fresh-wate puddles has also been recorded in rain-filled pools at Kline Point, Freetown (Muirhead Thomson, 1945), and accounts for the odd *melas* adults caught in nearby houses.

This remarkable breeding ground is entirely dependent on rainfall. After three weeks drought in the little dry season of August, it dried up in the middle of that month. The small rains started in the middle of September, and by the middle of October enormous numbers of larvae and pupae were again being found. This state of affairs continued till the beginning of December, when the compound dried up completely. The breeding conditions are closely mirrored in the fluctuations in numbers of *gambiae* caught in the houses (Table VI, fig. 1). From the separate records of these house catches it is seen that in such a congested area an output of 50,000 female *gambiae* per day produces a catch of not more than two or three *gambiae* per bedroom in adjacent houses. Or if it is put in another way a room index of only 1 in such a crowded area may still indicate that a considerable amount of breeding is taking place nearby.

Only one other breeding ground anything like this has been found in the crowded western part of Lagos Island, namely at Maloney Bridge Street on the opposite side of the island. This was a marshy patch of *Paspalum*, no longer connected with the sea, and containing several crab holes concealed among the grass, and flooded with fresh water. Of 10 eggs found all were *A. gambiae*. From the number of larvae and pupae found it was evidently a major breeding-place.

The practical lessons from these findings must be obvious. Failure to appreciate the fact that a whole corner of the city may be affected by the enormous *gambiae* output from a single compound, and failure to ensure that such breeding-places are not overlooked by the sanitary squads, may completely nullify all the good work being done elsewhere. The presence of such a limited number of breeding-places on Lagos Island points to the possibility that with very little extra effort the town of Lagos could be almost completely freed of *Anopheles*.

Maps of Lagos Island 40 years ago, and reference to reports at that time (Ross and others, 1902) show that *Anopheles* breeding on the island and its numerous swampy patches must have been very much more extensive in those days, and that there were probably many places resembling the Johnson St. compound. The present freedom of the greater part of Lagos Island from *gambiae* breeding-places is almost certainly due to the slow and inconspicuous but steady progress in street drainage and swamp filling over a period of years.

Summary.

The differences between the eggs of *A. gambiae* and *A. melas* form an absolutely reliable diagnostic character for distinguishing the two species in the Lagos area.

Identification of eggs laid by wild-caught females has formed the basis of work on the seasonal incidence of *gambiae* and *melas* adults in houses in different localities in and around Lagos.

In the area as a whole both *melas* and *gambiae* are abundant in most localities, but the greater part of Lagos town on Lagos Island is an almost pure *gambiae* area.

The sporozoite rate of *gambiae*—10·0 per cent.—is consistently much higher than that of *melas*—3·5 per cent.—in all catching stations. Fluctuations in sporozoite rate in relation to population density have been studied in both species.

The composition of the mosquito population in houses, and in outside resting places has been studied in relation to the movements of mosquitos into and out of houses. *Melas* makes use of outside resting places to about ten times the extent of *gambiae*.

In African village houses about 90 per cent. of the blood feeding by these two species takes place after midnight, the peak of activity being the hour or two before dawn.

An improved design of experimental hut and window trap is described. It has been used in studying the exodus of mosquitos from houses after feeding, and on experiments on host selection by *Anopheles*.

Swarms of male "*gambiae*" in nature have been described. In captivity both *melas* and *gambiae* have been persuaded to mate by using artificial light.

By cross fertilisation between *melas* and *gambiae* a generation of healthy hybrid adults can be obtained. Of these the females are evidently normal sexually, while most if not all of the males are sterile.

The breeding places of *melas* and *gambiae* in and around Lagos have been studied in relation to control. In this lagoon area, where conditions are very different from those of Freetown Estuary, *melas* breeds under a variety of conditions, and *Avicennia* mangrove is only of limited value in indicating major breeding grounds.

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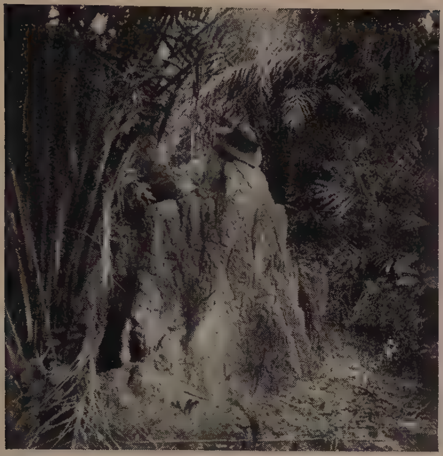

Fig. 1. Shaded termite mound in palm thicket. Anjorin, near Lagos. Outside resting place of *melas*.

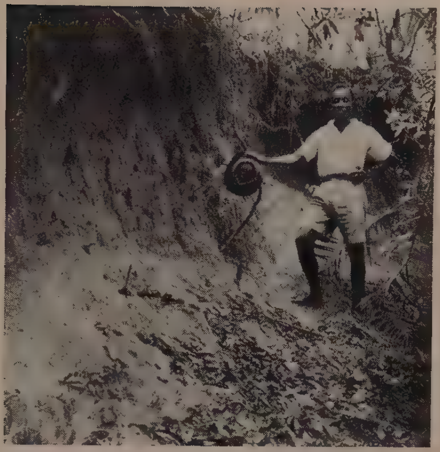

Fig. 2. Heavily-shaded earth bank. Makoko, near Lagos. Regular outside resting place of *A. melas* and *A. gambiae*.

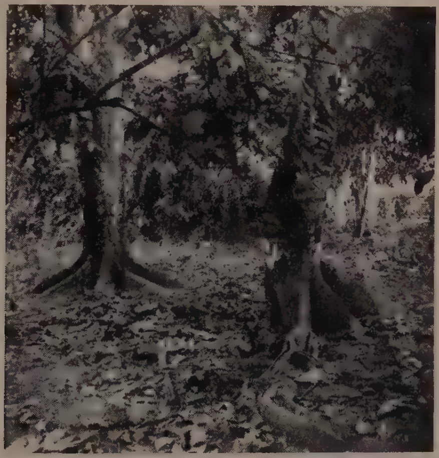

Fig. 3. Heavily-shaded bases of forest trees. Senu, near Lagos. Occasional outside resting place of *A. melas* and possibly *gambiae*.

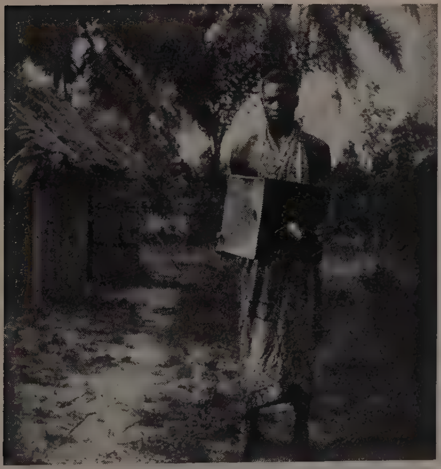

Fig. 4. Detachable window-trap cage used in experiments on normal exodus of mosquitos from houses. Also used in studying behaviour of mosquitos in houses treated with Pyrethrum and with DDT in Kerosene.

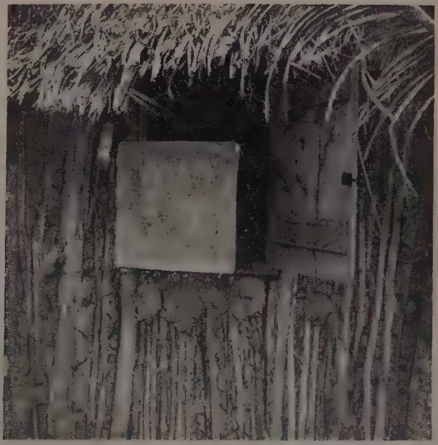

Fig. 5. Window-trap cage fixed over window of experimental hut.

FIELD OBSERVATIONS ON THE BEAN SEED FLY (SEED CORN MAGGOT), *CHORTOPHILA CILICRURA*, ROND., AND *C. TRICHODACTYLA*, ROND.

By Mary Miles.
Long Ashton Research Station, University of Bristol.

In the market gardening areas of south Worcestershire, the heavy annual loss to germinating beans through attacks by larvae of *Chortophila cilicrura*, Rond., the bean seed fly (seed corn maggot of U.S.A.) has created a demand for control measures, but owing to lack of knowledge of the biology and habits of the flies none could be devised. In Britain the larvae were known to feed in beans in the latter part of May and early June and give rise to flies towards the end of June, but from then until the injury occurred again on beans in the following spring nothing was known of the behaviour of the species. The field observations described here were undertaken to obtain information on the habits and food-plants of *C. cilicrura* throughout the year and it was hoped that such information would provide a basis for control measures.

Distribution.

The occasional occurrence of injury by bean seed fly was noted by Smith (1931) who drew attention to the existence of this potential pest in widely separated localities in Britain. Since 1931 the Monthly Summary of Insect and Allied Pests occurring in England and Wales (Ministry of Agriculture) has recorded attack by *C. cilicrura* almost every spring, and the sites of the attacks have shown that the species is widely distributed. In 1932 injury by *C. cilicrura* was recorded from Devonshire; in 1933 it was reported from North Wales; in 1935 from Surrey and the west of England; in 1936 from Devon and Cornwall; in 1937 from Worcestershire; in 1938 from Bristol, Worcestershire and Hampshire; in 1939 from Worcestershire and Gloucestershire; in 1940 from Manchester, Reading, Bristol, Wiltshire, Worcestershire, Somerset and Cheshire; in 1941 from Cheshire, north Lancashire, Hampshire, Worcestershire, and Devonshire; in 1945 from Surrey and other parts of the south-east of England, Birmingham and Shropshire; and in 1946 from Devon and Sussex.

References to *C. cilicrura* in the *Review of Applied Entomology* show that the insect is widely distributed throughout the temperate zones and that it is of considerable economic importance in Japan and Manchuria and throughout eastern and western Europe, North America and the West Indies and parts of South America.

Species concerned in Attacks on Crops in Britain.

The rearing of larvae from germinating beans has shown that two species, *C. cilicrura* and *C. trichodactyla*, Rond., are associated in these attacks, and both species have been bred from leeks and spring cabbages. Examination of living larvae has not so far revealed characters by which the larvae of the two species can be distinguished. In the adult stage males of the two species are distinct but females are indistinguishable. In view of the close association of *C. cilicrura* and *C. trichodactyla* in the field, the names *C. cilicrura* and bean seed fly are used in this paper for both species except in sections relating to their relative prevalence and distinguishing characters.

The comparison of the prevalence of the two species is based on the numbers of males, and data so far collected suggest that in south Worcestershire *C. cilicrura* is more numerous than *C. trichodactyla*. Of 75 males bred or captured in the course of these observations, 52 were *C. cilicrura* and 23 were *C. trichodactyla.* From one collection of larvae from beans, 26 males were reared and of these 20 were *C. cilicrura* and 6 were *C. trichodactyla.* These numbers are not considered sufficient to show reliably the relative importance of the two species in south Worcestershire since bias may have been introduced during collecting by such factors as time of day, weather conditions, date, crop and season.

The proportion of males and females among the bred and captured flies is of interest. In a batch of 53 flies bred from a single collection of larvae, 26 were males and 27 were females, which suggests that the numbers of males and females produced may be approximately equal. In a total of 177 adults bred and collected in the course of these observations, 75 were males and 102 were females. It is recognised, however, that differences in the habits of males and females may affect the proportion of the sexes among captured insects for Hawley (1922) observed that females seemed to be more numerous than males on bare soil and males seemed to predominate about herbage.

Description of the Species.

Adults.

The following description of the adults is not complete but it should assist in the recognition of the flies and the separation of the males of the two species.

The flies are 4–5 mm. in length. Males are dark greyish brown with the thorax more or less distinctly striped dorsally ; the abdomen has a mid-dorsal stripe and faint transverse stripes at the edges of the segments, and an inconspicuous hypopygium. Females are much lighter and more yellowish than males and the dark stripes may be absent or only faintly indicated. In both sexes the legs are dark. Wings are 4–4·5 mm. in length and are transparent grey without any trace of yellow on the membrane ; the squamae are approximately equal in size and the veins are greyish brown. The costa has short inconspicuous spines and extends along the margin of the wing to the medius ; the medius is almost straight, and the first anal vein extends to the wing margin.

The head is greyish brown, dark in the males and light in the females. In the males the eyes are narrowly separated, but in the females they are separated by a light greyish band about one-third of the width of the head and a reddish area occurs above the lunule. The eyes are without microscopic hairs ; the arista is sparsely pubescent ; the proboscis is slender, and the palpi are dark.

The thorax is rather narrower than the head and is convex dorsally. Distinctive features of the chaetotaxy are found in the relative lengths of the first (pre-alary) and second supra-alary bristles and of the upper and lower hind sternopleural bristles. In males of *C. cilicrura* the first supra-alary bristle is rather less than half the length of the second, while in males of *C. trichodactyla* the first supra-alary bristle is very much smaller than the second. In both species the lower hind sternopleural bristle is shorter by about a quarter of its length than the upper hind sternopleural bristle. In both species the scutellum is unicolorous or darker dorsally.

Males of *C. cilicrura* and *C. trichodactyla* differ in the chaetotaxy of the legs. In males of *C. cilicrura* the meta-thoracic femora have 3–4 long postero-ventral bristles near the apex, and the metathoracic tibiae have a comb-like row, often double at the base, of long, slender, down-curved bristles postero-ventrally along the whole length, and 8–9 long antero-dorsal bristles ; the mesothoracic metatarsi have short bristles on the dorsal face and often a few longer stouter bristles also. In males of *C. trichodactyla* the metathoracic femora have a more or less complete row of postero-ventral bristles and the metathoracic tibiae have a comb-like row of bristles shorter and less curved than those of *C. cilicrura* and the antero-dorsal bristles are less numerous and regular; the mesothoracic meta-tarsi have a dorsal band of long fine bristles directed towards the apex at such an acute angle that they often lie close to the surface of the segment.

Immature stages.

Egg.—Eggs of *C. cilicrura* are about 1 mm. long, narrowly elliptical and glistening white with the surface sculptured with broken longitudinal ridges. This description is equally applicable to eggs of other root-feeding Anthomyiids, and it is apparent that a comparative study of eggs of Anthomyiids will be necessary before specific characters can be distinguished.

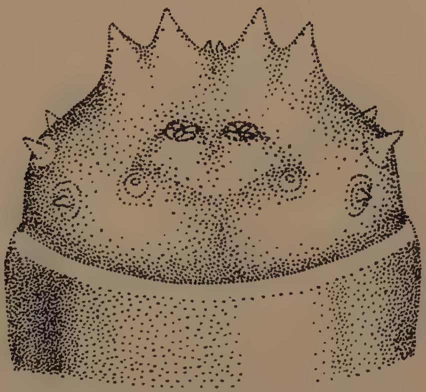

Fig. 1.—Caudal segment of larva of *C. cilicrura*, Rond. Viewed dorsally (×42).

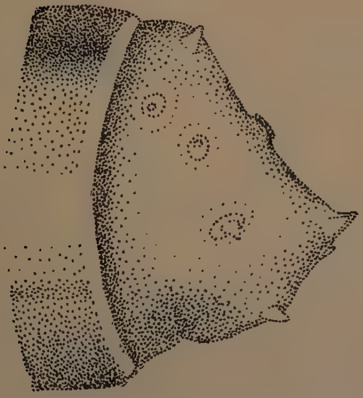

Fig. 2.—Caudal segment of larva of *C. cilicrura*, Rond., in profile (×42).

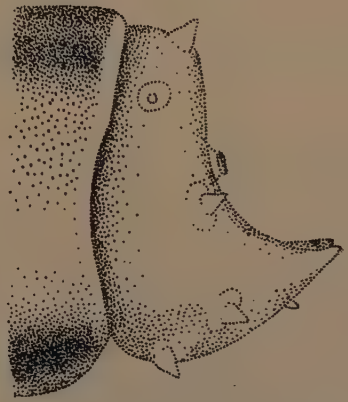

Fig. 3.—Caudal segment of larva of *H. antiqua*, Mg., in profile (×33).

Larva.—A complete description of the larva of *C. cilicrura* is not yet available because it is hoped that further investigation will reveal differences between larvae of *C. cilicrura* and *C. trichodactyla*. The resemblance between larvae of various Anthomyiids makes it advisable to include in a description of *C. cilicrura* some comparison with other species so that distinctive features receive proper emphasis, but at present this cannot be done adequately because it has not been possible to examine larvae of a sufficiently wide range of species. The following notes, however, should be of assistance in the recognition of larvae of *C. cilicrura*:—

The larvae are white, slender, rather smoothly rounded, and with the segmentation distinct but not emphasised by transverse wrinkling of the integument. The head is vestigial, the mouth parts are reduced to heavily chitinised hooks, legs are absent and the body is thickest at the hind end. The caudal segment is slightly drawn out posteriorly and flattened dorso-ventrally so that in profile it is somewhat triangular. The posterior spiracles are situated on small tubercles arising from the flattened dorsal surface, and the anus is visible as a median longitudinal slit on the ventral surface near the base. The caudal segment also bears 8 pairs of fleshy tubercles, 5 pairs of which form the corona or ornamentation of the flattened dorsal surface and three pairs occur on the ventral surface. Figure 1 shows the relative position of the 5 pairs of dorsal tubercles and the tips of the ventro-apical and ventro-lateral pairs are also visible ; the third pair of ventral tubercles flank the caudal end of the anus. Fully-grown larvae are 5·5–6·5 mm. long.

Preliminary observations suggest that the character and arrangement of the tubercles of the posterior segment may be of value in the identification of root-feeding species of Anthomyiids. Larvae of *C. cilicrura* are readily distinguished from those of *C. brassicae*, Bch., the cabbage root fly, by the character of the apical tubercles which are simple in *C. cilicrura* and bifid in *C. brassicae*. The same character separates the larvae of *C. cilicrura* from those of *Hylemyia coarctata*, Fall., the wheat bulb fly. The separation of larvae of *C. cilicrura* from those of *Hylemyia antiqua*, Mg., the onion fly, is more difficult. The shape and arrangement of the tubercles forming the corona are similar in both species, but in *H. antiqua* the tubercles are relatively larger and more conspicuous. A comparison of figs. 2 and 3 show that the caudal segment of *H. antiqua* is relatively narrower and more truncate than that of *C. cilicrura*, and in *H. antiqua* the anal tubercles are much nearer the base of the segment. Fully-grown larvae of *H. antiqua* are much larger and stouter than those of *C. cilicrura*, the former being 9–10 mm. in length while the latter do not exceed 7 mm.

Puparium.—The puparium is brown or dark brown, elongate oval, 4–5 mm. in length and about 1·25 mm. in breadth. The posterior tubercles characteristic of the larva are recognisable on the puparium and can be used to identify it. Puparia of *C. cilicrura* are smaller than those of *H. antiqua* and the posterior projections (remains of larval tubercles) are also smaller and less conspicuous.

Food-plants of *C. cilicrura* in Britain.

Beans.

C. cilicrura, the bean seed fly, is frequently found at the roots of beans. Attack takes place before the seedlings are through the soil and the maggots are generally about fully grown before infestation is discovered. They penetrate the cotyledons as soon as the testa splits and feed in tunnels in the interior or in shallow winding depressions beneath the testa. The tunnels become full of wet brownish rotting matter that keeps the maggots bathed in moisture. In healthy cotyledons the rot does not extend beyond the feeding tunnels but when diseased seeds are attacked the cotyledons speedily decay.

TABLE I.

Plot	No. of seedlings examined	Rotting beans		Healthy beans	
		Attacked	Not attacked	Attacked	Not attacked
A	91	4	8	64	15
B	99	1	14	48	36
X	105	27	8	62	8
Y	101	35	2	62	2

In America it was thought (Bonde 1939) that larvae of *C. cilicrura* were attracted to diseased potato sets, and in consequence an attempt was made to discover if they showed a preference for diseased seed beans. Random samples of approximately 100 " misses " and stunted seedlings from each of 4 plots were examined and the results are given in Table I.

The data collected gave no evidence that the larvae preferred diseased seeds. The proportion of healthy seeds attacked by maggots was greater than that of diseased seeds, but the reason for this appeared to be that healthy seeds made better growth than diseased seeds and thus exposed a greater area of plant tissue to invasion.

The feeding of a few (<3) larvae in healthy cotyledons had little perceptible effect on growth but when several maggots were present the food reserves of the seedlings were seriously depleted. Injury to the embryonic region prevented germination, and seedlings were killed or greatly weakened when attack by maggots aided the spread of fungi and bacteria in the cotyledons. When dwarf beans were attacked the cotyledons were often pushed through the soil with maggots feeding in the tissue and their feeding tunnels showed later as brown scars and deformities. In runner beans the cotyledons remain in the soil and maggots may be found feeding in them after the seedlings are established.

Penetration of hypocotyls of dwarf beans and epicotyls of runner beans by larvae of *C. cilicrura* was often fatal to the seedlings. Upward tunnelling in runner beans caused the first pair of leaves to be deformed or suppressed and the terminal bud to be killed, and many attacked seedlings appeared above ground as short leafless stems. Upward tunnelling in the hypocotyls of dwarf beans often destroyed the plumule and made the seedlings blind and snake-headed. Downward tunnelling tended to sever the cotyledons from the radicle and often checked root development. Occasionally the maggots fed superficially on epicotyls and hypocotyls and caused the seedlings to be stunted and deformed. Injury to the shoot was often followed by soft rot and the collapse and death of the seedlings. Loss of the terminal shoot was sometimes followed by the development of lateral shoots but no data were collected on their market value.

Peas.

Attack on peas by *C. cilicrura* is less common than attack on beans, probably because peas are usually sown earlier than beans and the plants are established before the spring generation of *C. cilicrura* is active. When germinating peas are attacked the maggots feed in the cotyledons and shoots.

Spring Cabbages.

In October 1945 larvae of *C. cilicrura* were found to be widely distributed at the roots of a crop of newly planted spring cabbages. Hitherto the only recorded root-feeding maggot on cabbages and allied crops in Britain has been the cabbage root fly, *C. brassicae*, but in the crop under observation no larvae of this species were found. *C. cilicrura* has been recorded as a pest of cabbages in U.S.A. (Fulton 1942) and its discovery at the roots of cabbages in Britain was an important contribution to the knowledge of its habits.

Examination of a random selection of plants showed that infestation was general and that 1–4 larvae were present in attacked plants. The crop showed no obvious signs of attack during the autumn but when examined again the following spring a large number of " bolters " and plants with soft rot were found. No relationship between autumn attack by *C. cilicrura* and the occurrence of bolting and soft rot in spring was established but the subject needs further investigation. Blackened areas near the base of the stem of some bolted plants were thought to be the site of insect attack, possibly attack by *C. cilicrura*, at a much earlier stage in their growth, and bolting might have followed the check to growth resulting from the injury. Plants with soft rot were not examined critically but *C. cilicrura* has been shown to be a vector of bacterial diseases (Leach 1940), and the possible association between attack by *C. cilicrura* in autumn and the incidence of soft rot in spring cannot be ignored.

Other Brassicae.

Larvae of *C. cilicrura* were found during spring and summer in company with larvae of *C. brassicae* at the roots of cauliflowers, summer cabbages, brussels sprouts and kale at various places in the west of England. At that time of the year *C. brassicae* was the dominant species, and when present *C. cilicrura* constituted less than 10 per cent. of the root-maggot population.

Leeks.

Leeks set out in June and July were attacked by larvae of *C. cilicrura* during July and infested plants contained 1–5 maggots in the roots. It was not possible to recognise infested plants in the field but examination of random selections of leeks showed the larvae to be widely distributed in the two crops under observation.

Onions.

C. cilicrura was bred from cultures of maggots taken at the roots of onions. The onion fly, *Hylemyia antiqua*, was the dominant species on the onions examined but the occurrence of *C. cilicrura* in association with *H. antiqua* was evidence that onions were among the food-plants of *C. cilicrura* in Britain.

Cucumbers.

In June 1946 larvae of *C. cilicrura* tunnelled in the hypocotyls of seedling cucumbers growing out of doors near Evesham, and injury was so severe that the crop was ploughed in. Similar injury has been observed by other workers but the insect associated with it was not identified. In June 1944 Staniland recorded (Min. Agric. Mon. Rep.) unidentified Dipterous larvae tunnelling in hypocotyls of ridge cucumbers at Penzance, and in May 1946 Petherbridge recorded from the eastern counties (Min. Agric. Mon. Rep.) larvae, ? *C. cilicrura*, boring in hypocotyls of cucumbers in the cotyledon stage. *C. cilicrura* is a pest of CUCURBITACEAE in U.S.A. (Hawley 1922) and West Indies (Ogilvie 1928).

Lettuce.

In December 1944 a larva of *C. cilicrura* was taken out of doors in the root of a lettuce. The infestation by *C. cilicrura* occurred simultaneously with severe attack by larvae of mud beetles (*Helophorus* sp.).

Biology and Annual Cycle of *C. cilicrura.*

The study of the biology and habits of *C. cilicrura* present some difficulty because the flies do not mate readily in captivity and the resemblance of the various stages to those of other Anthomyiid flies makes accurate identification in the field almost impossible. In spite of this, studies of *C. cilicrura* have been carried out in North America, Japan and Russia, and much biological information has been collected.

The economic importance of *C. cilicrura* on seedlings of maize and beans and in cut sets of potato led to studies of the spring generation in U.S.A. where the flies are active from mid-May to mid-June (Hawley 1922, Bonde 1939). After emergence from the puparia the flies have a pre-oviposition period which appears to vary in length with the season (Harukawa and others 1930 & 1934, Rekach 1932). They are most active in sunshine and lay eggs in moist, freshly turned soil (Bonde 1939). Hatching takes place after an incubation period of 1–8 days (Reid 1940). The larvae feed in vegetable matter and reach maturity in 7–12 days (Bonde 1939), but at low temperatures the feeding period is greatly extended (Harukawa and others 1934, Reid 1940). The pupal stage is passed in the soil and lasts 7–18 days in spring (Reid 1940) but extends for some months during winter and during diapause (Rekach 1932).

The summer generation of *C. cilicrura* has received little attention, probably because it has not been directly associated with severe injury to crops. Hawley (1922) found that flies of second generation frequented grasses and cereals and suggested that these crops might be alternate hosts for later generations, but as he seldom saw the flies during the summer he concluded that the hot weather of mid-July was fatal to them. In Russia Rekach (1932) noted the absence of the flies during the summer months ; Harukawa and others (1933a) found that few of the flies came to baits during the period July to September ; and Reid (1940) recorded that flies were least abundant at mid-summer.

All these observers state that the flies are again numerous in autumn and it would appear, therefore, that the scarcity of *C. cilicrura* during the summer months is more apparent than real. There is some evidence in recent literature to support this view. Vasil'ev (1936) found that there were two generations of *C. cilicrura* and *C. trichodactyla* in the course of spring and summer, and lupins for fodder sown at different dates were attacked by the larvae. Schwardt (1943) also observed that adults of *C. cilicrura* were scarce or absent after mid-July but found that plots planted on 31st July and examined 11th August showed 40 per cent. damage by the larvae. In their account of attack by larvae of *C. cilicrura* on seedlings of douglas fir and spruce, Breakey and others (1945) appear to be dealing with the summer generation of larvae as the tree seeds had been sown on 25th June and injury had occurred between date of planting and 21st July when most of the maggots had finished feeding ; and after completing their development the flies emerged at the end of July and the beginning of August, the expected time for the third generation.

The autumn generation of *C. cilicrura* is widely known in France as a pest of winter cereals (Balachowsky & Mesnil 1935, Couturier 1937). Adults are abundant in autumn in Japan (Harukawa 1930, 1933a) and in South Russia (Rekach 1932). Reid (1940) records that flies are common in late autumn on the Atlantic coastal plain of southern U.S.A. where the larvae are a serious pest of spinach sown between 1st October and 15th November. Schwardt (1943) records from New York the occurrence of larvae of *C. cilicrura* in October.

Some evidence of activity by *C. cilicrura* throughout the winter months has been collected. Harukawa (1930) recorded that flies emerged in January and that even in cold winters a few flies were active in the warmer part of the day. Rekach observed (1932) that hibernation occurred in larval, adult and pupal stages and that overwintering flies lived 4–5 months. Reid (1940) stated that *C. cilicrura* appeared to breed all the year round in North Carolina.

Field Observations of *C. cilicrura* in the West of England.

Most of the observations recorded here were made in the Evesham area of south Worcestershire on a large arable farm devoted mainly to vegetable crops. A two-year rotation was followed and much of the land carried two crops a year. The soil was typical of the Pershore series, a coarse, heavy sand of considerable depth, with drainage slightly impeded at a depth of about 2 ft. The annual rainfall was about 25 inches and the district was noted for its sunny sheltered situation. The farm was highly mechanised, and hoeing and transplanting were generally carried out with machines. The crops left a good deal of exposed soil and the rapid growth of weeds involved almost continuous cultivation between the rows. Overhead irrigation was used freely in spring and summer to hasten the establishment of transplanted crops and to moisten the soil of seed beds so that seedlings could be lifted easily without injury to their roots. At the time of the observations the manurial practice had been modified by war-time shortages of fertilisers ; but stock were kept on the farm and dung and liquid manure were available, and these were supplemented by such fertilisers as were obtainable.

The records of the occurrence of *C. cilicrura* have been given in a seasonal sequence, and observations made during 1945–46 have been supplemented by reference to records obtained in previous years. It was convenient to begin the cycle in autumn.

Autumn and winter observations.

In 1945 adults of *C. cilicrura* were numerous from mid-September to mid-October during periods of warm sunny weather. They were captured on spring cabbages set out in mid-September and on bare soil after it had been ploughed on 5th October. Flies captured 8th October contained fully developed eggs and a female captured 9th October laid eggs during 9–14th October. On 11th October eggs that appeared to be those of *C. cilicrura* were recovered from the soil of a field ploughed a week previously but their identity could not be established because they had been damaged during their extraction from the soil ; larvae of *C. cilicrura,* however, were found in this field a fortnight later.

On 19th October a larva of *C. cilicrura* was found in a bean seed that formed part of the remains of a bean crop that had been buried by ploughing prior to planting spring cabbages. The testa of the bean was ruptured and the larva had entered the seed near the micropyle. It fed in the cotyledons until it reached maturity. During 26–30th October it burrowed into soil for pupation and the fly emerged in the laboratory on 9th November.

In the meantime 6 eggs laid 9–14th October in a glass phial hatched. The larvae were offered slices of petiole of cabbage into which they tunnelled. The tube containing the larvae was kept at room temperature and soft rot quickly developed in the cabbage tissue. The larvae continued to feed in healthy and rotting cabbage stem tissue until they reached maturity and entered the soil for pupation during 26th October–4th November.

On 26th October larvae of *C. cilicrura* were found in numbers in the stems and petioles of partly buried cabbage seedlings from a seed bed ploughed on 5th October. The larvae were nearly fully grown when found and at the time of infestation the buried seedlings showed little sign of decomposition.

On 29th October larvae of *C. cilicrura* were found in the stems of spring cabbages that had been planted in mid-September. By this time the larvae were nearly mature but they were still present in the crop in early November when field observations ceased.

The last recorded date of emergence of *C. cilicrura* out of doors in 1945 was 11th October. The larva had been collected at cauliflower on 15th September.

Larvae of *C. cilicrura* collected in October and early November 1945 and kept at room temperatures throughout the period had a pupal stage of 12 to 18 days and emerged as flies from 9th November until early December. A few of the pupae entered a diapause and emerged as adults in February and March.

In 1944 some evidence was collected of activity of larvae and adults of *C. cilicrura* during late autumn and winter. Larvae collected on 19th September were placed in an outdoor insectary and during 9–20th November 8 flies (♂♂ and ♀♀) emerged. They were offered sugar solution and meat extract (oxo) and were caged over soil in which beans were planted. By 4th December only 4 flies remained alive, but they were active in sunshine until February although there were severe frosts and snow in January which blackened the bean seedlings in the cage. During 15–21st February the over-wintering flies died and no eggs or larvae were found in the cage.

This late activity of *C. cilicrura* in captivity did not appear to be fortuitous. On 7th December of the same year a larva was found in the root of a lettuce plant set out in November. The presence of an active larva in December suggested that adults had been on the wing in late autumn when the ground was prepared for the crop.

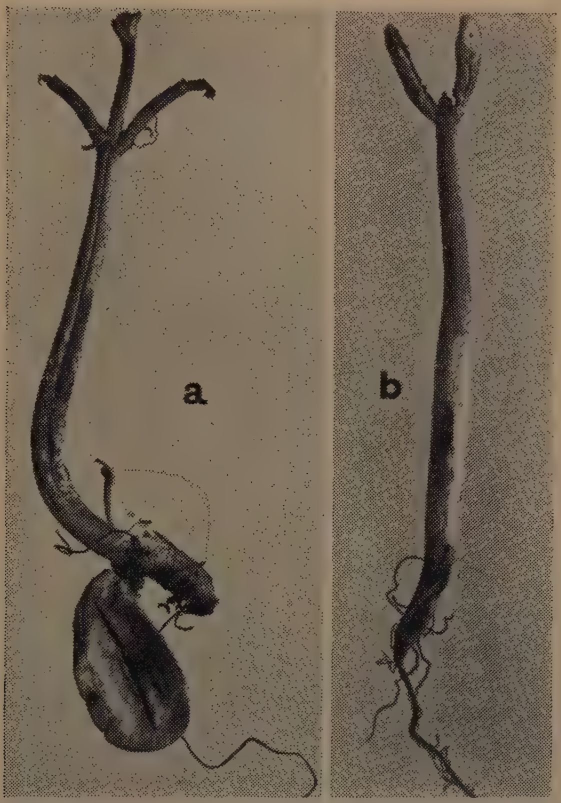

Fig. 4. (*a*) Seedling of runner bean showing injury by larvae of *C. cilicrura*, Rond., to cotyledons, hypocotyl and epicotyl. There is almost complete loss of radicle and the leaflets are aborted. (*b*) Seedling of dwarf bean showing injury by larvae of *C. cilicrura*, Rond., to cotyledons, hypocotyl and radicle and the abortion of the terminal bud.

Spring observations.

In 1945 adults of *C. cilicrura* emerged from hibernating puparia from 1st March to 17th April and the first adult was taken on the wing on 26th March. In 1946 the first adult was captured on 9th April. In both 1945 and 1946 *C. cilicrura* was common near Evesham from the second week of April until mid-May and on sunny days the flies settled in small flocks on patches of freshly raked soil.

No observations have been made on the period between the emergence of the flies and egg-laying, but flies frequented the bean beds when planting was in progress in the middle of April. A fly captured on the bare soil of a bean bed on 16th April laid eggs in captivity 19–21st April.

In 1946 larvae were first taken in germinating runner beans on 13th May and from then until the end of the first week of June they were readily found in the cotyledons and epicotyls of bean seedlings. Larvae from beans began to enter the soil for pupation on 25th May and by 10th June few larvae remained at the beans.

A batch of 6 larvae that entered the soil 25–26th May emerged 17–19th June. Mass cultures of larvae from beans pupated 4–10th June and emerged 25th June–3rd July with a peak of emergence during 26–29th June.

Larvae of *C. cilicrura* were taken in germinating seeds of ridge cucumbers on 9th June. By this time many seedlings had been killed and larvae found were about fully grown.

Summer observations.

The second generation of *C. cilicrura* was on the wing from mid-June to early July and the flies were found on beans, carrots, Brassicae, on the bare soil between rows of newly hoed carrots, on soil being prepared for leeks and on young leeks after they had been transplanted from the seed beds.

On 9th July roots of leeks selected at random were examined and larvae of *C. cilicrura* were found. They reached maturity on the leeks, entered the soil for pupation on 18th July and emerged 3–7th August.

On 23rd July a later crop of leeks was examined and larvae of *C. cilicrura* were again found in the roots. They were mature and entered the soil shortly after being captured. They emerged 13–15th August.

During the latter part of June and in July larvae of *C. cilicrura* were also taken at the roots of brussels sprouts, cabbages, cauliflowers, kale and dwarf beans.

No field observations were made during August. The examination of carrot seedlings was planned because flies had often been seen on carrots but wet weather postponed examination until 9th September, a date too long after the preparation of the seed bed and the disturbance of the soil by harrowing for larvae to be present. As was expected no larvae were found in soil samples from carrot rows but several empty pupal cases and two viable pupae were found. This cannot be considered evidence that larvae of *C. cilicrura* feed on carrots but it suggests that in the case of carrots further search should be made.

Annual Cycle of *C. cilicrura* in Britain.

From data already assembled it is possible to reconstruct the annual cycle of generations of *C. cilicrura* in Britain. Flies have been taken on the wing every month from March to October and there is some evidence of slight activity during the winter. There appear to be 3–4 generations a year which corresponds with the observations of Rekach (1932) in Russia and Schwardt (1944) who noted in New York that there were at least two generations after the spring generation. In Britain flies of the first generation are active from mid-April to mid-May ; their larvae feed in germinating seeds of beans and cucumbers and at the roots of summer cauliflowers. Flies of the second generation are on the wing in the latter half of June and early July and have been captured on beans, various Brassicae, leeks, carrots and headland herbage ; their larvae have been found at the roots of Brassicae and leeks. The third generation of flies is on the wing during the first half of August and their larvae feed at the roots of transplanted spring cabbages. The fourth generation is on the wing from mid-September to mid-October and gives rise to larvae that have been taken at the roots of late-transplanted spring cabbages and lettuce. There appears to be some overlapping of the generations, particularly in autumn, and the flies may be numerous in sunny weather from August to October.

The following dates of finding immature stages of *C. cilicrura* show that larvae are active from May to December. Peak periods of larval activity occur during late May and early June, in July, and during the period August to October. The spring peak of activity on beans has been widely recognized but the summer and autumn generations of larvae have hitherto passed unnoticed in Britain.

TABLE II.

Date	Crop	Locality	Stage
13.v.46–4.vi.46	Beans	Evesham	Larvae
9.vi.46	Cucumbers	Evesham	Larvae
22.vi.46	Brussels sprouts	Yeovil	Larvae
25.vi.46	Cauliflowers	Evesham	Pupae
4.vii.46	Cabbages Brussels sprouts Dwarf beans	Bromham	Larvae and pupae
9.vii.46	Leeks	Evesham	Larvae
19.vii.46	Kale	Chippenham	Larvae
23.vii.46	Leeks	Evesham	Larvae
9.ix.46	Carrots	Evesham	Pupae
15.ix.46	Cauliflowers	Evesham	Larvae
19.ix.44	Spring cabbages	Evesham	Larvae
19.x.45	Bean crop residue (buried)	Evesham	Larvae
26.x.45–6.xi.45	Spring cabbages Cabbage seedlings (buried residues)	Evesham	Larvae

Association between *C. cilicrura* **and rotting Vegetation.**

American investigators have drawn attention to an apparent association between the presence of rotting vegetation and the occurrence of *C. cilicrura*. Hawley (1922) recorded that *C. cilicrura* was strongly attracted to moist and decaying vegetation for egg-laying and that eggs had been found around decaying bean haulms and rotting cabbages; and Bonde (1939) found that cut sets of potatoes affected by disease were more frequently attacked by *C. cilicrura* than healthy sets. In the course of observations in Worcestershire any association of maggots of *C. cilicrura* with decaying vegetation was noted.

Observations so far made suggest that there is no direct association between the occurrence of *C. cilicrura* and the presence of decaying vegetation. It has already been shown that *C. cilicrura* showed no preference for diseased bean seeds, and soft rot in hypocotyls and epicotyls appears to be a result, and not a condition, of attack. Transplanted cabbages and leeks were healthy when attacked and decomposition had not started in buried crop residues of beans and cabbages when larvae of *C. cilicrura* attacked them.

A search was made among rotting plants for larvae of *C. cilicrura*. In September 1945 a plot of swedes with extensive heart rot was examined. Large numbers of *Phaonia trimaculata*, Bouché, were found in the rotting tissue together with occasional larvae of *C. brassicae* but no larvae of *C. cilicrura*. The remains of an adjoining cabbage crop were also examined. Larvae of *C. fugax*, Meig., were found in the rotting stems but no larvae of *C. cilicrura* were present.

In May 1946 spring cabbages that had been infested with *C. cilicrura* in the preceding autumn were examined. Many of the plants had rotting leaves and adults of *C. cilicrura* were numerous on the crop. Eggs and larvae of *C. fugax* were present in large numbers on the rotting leaves but no larvae of *C. cilicrura* were found.

In July 1946 the remains of a cauliflower crop were examined. The small rejected heads were beginning to rot and insect eggs were found near the rotting tissue. Larvae from the eggs fed on the rotting plants and when flies emerged they were identified as *Muscina assimilis*, Fall. In August 1946 *M. assimilis* was bred from rotting heads of cauliflower collected in Herefordshire.

Cultural Practice and the Incidence of *C. cilicrura*.

Observations on *C. cilicrura* suggest that it occurs as a pest in market gardens when cultural operations are carried out during periods of fly activity. In the

Evesham area there is a succession of short-term crops with extensive areas of bare soil between the rows and almost continuous surface cultivation to keep down weeds and conserve moisture, and thus conditions attractive to the egg-laying flies are maintained throughout the season. The system of culture also provides favourable conditions for the survival and development of the larvae, for food plants are almost always present and buried crop residues and organic fertilisers are available to supplement larval diet and to maintain suitable soil conditions.

A noteworthy feature concerning the incidence of *C. cilicrura* is that the higher the standard of cultivation the more favourable the conditions are for the development of the flies. Irrigation, intended to assist the rapid establishment of seedlings and transplanted crops, maintains the attractiveness of the soil for the egg-laying females for long periods after cultivation, and provides adequate moisture to ensure the hatching of eggs and the survival of larvae until they reach food. It also masks the signs of insect attack on crops and enables them to support higher populations of injurious insects than would otherwise be possible. The writer has seen summer cauliflowers that were heavily infested with *C. brassicae* and *C. cilicrura* established by means of overhead watering and then ploughed in at harvest because, owing to insect attack, the heads were too small and stunted for market.

Climatic conditions favourable for intensive vegetable culture also favour *C. cilicrura* for the flies are most active in the sunny sheltered situations that are sought for the cultivation of vegetables. How far soil conditions affect *C. cilicrura* has not been investigated but it has been observed that the flies are less numerous about vegetable plots at Long Ashton Research Station where the soil is heavy than in the Evesham area where light soils prevail.

Natural Enemies of *C. cilicrura.*

Parasites of *C. cilicrura* in south Worcestershire include a small Staphylinid beetle, *Aleochara bipustulata*, L., which was identified by Dr. A. M. Massee of East Malling Research Station, and a Hymenopterous parasite, *Trybliographa diaphana*, Hartig, which was identified by Mr. G. J. Kerrich of the Manchester Museum. Predatory maggots have been found in beans along with larvae of *C. cilicrura* and the tiger fly, *Coenosia tigrina*, F., has been reared. Larvae of LEPTIDAE have also been found in beans infested with *C. cilicrura.*

Control of *C. cilicrura.*

Experiments on the control of *C. cilicrura* have taken many forms and direct and indirect measures have been recommended.

Seed dressings.—Hawley (1922) recorded that gas tar, copperas, coal tar with ashes, lime and tobacco dust had been recommended as protective dressings for seeds, and he experimented with paraffin, lead arsenate, bordeaux mixture and lime sulphur with some measure of success. But substances that gave protection against attack by larvae of *C. cilicrura* tended to injure seeds and seedlings, and the splitting of the seed coat at germination and the growth of the seed left vital parts of the plant unprotected. Hawley (1922) and Glasgow (1934) concluded that seed treatments were not satisfactory for the control of *C. cilicrura* on a field scale.

Soil dressings.—Soil dressings have included repellents against adults and larvae, toxic dressings against larvae, artificial fertilisers and neutral substances like gypsum and ground limestone. Repellent and toxic substances are not always distinct and among those that have been tested are tarred sand, carbolic acid and lime, paraffin and lime, creosote and lime, creosote emulsion, ammonium sulphate, sulphur, bordeaux mixture, carbon bisulphide, carbolic emulsion, potassium cyanide, tobacco dust, chlorpicrin, and a mixture of dichloropropylene and dichlorpropane (D.D.). Some success has followed the use of these dressings (Hawley 1922, Harukawa 1933b, Wilford 1940, Breakey 1945) but many are injurious to the seedlings and others unsuited to use on a field scale. Artificial fertilisers such as calcium cyanamide, sulphate of potash, nitrate of soda, acid phosphate, nitrate of potash and dried blood have been used in the rows to protect the seeds from attack and to stimulate growth, but their application frequently resulted in the scorching of the seedlings (Hawley, 1922). Ground limestone and gypsum seem to have had no protective effects.

Baiting and trapping.—Rekach (1932) used a poison bait consisting of paris green and cotton seed meal (1 : 25) and obtained 30–48 per cent. mortality among the larvae by broadcasting it at the rate of 30 lb. per acre on the day previous to sowing seed and harrowing it into the soil. Hawley (1922) used a poison bait consisting of sodium arsenite, molasses and water against the flies but although he found dead flies at the bait there appeared to be no reduction in the number of attacked seedlings. Rhodium oil has proved attractive to *C. cilicrura* (Marshall, 1931), and in preliminary trials of attractants Mellor and Woodman (1933) found that *C. cilicrura* came to fly traps baited with yeast, allyl mustard oil and glycine. Rekach (1932) obtained promising results by the use of shallow pits (10 in. by 1 sq. yd.) baited with cotton seed meal and covered lightly with soil and kept damp ; the pits were distributed at the rate of about one per acre, the bait attracted the egg-laying flies and concentrated the larvae, which were killed after three weeks with sodium arsenite.

Indirect methods of control.—Much can be done to reduce attack by *C. cilicrura* by cultural means, and the following measures have been recommended : the preparation of seed beds when flies are not on the wing (Rekach 1932, Wilford 1940), delaying seeding until the egg-laying period is over or until larvae have finished feeding (Hawley 1922, Engel'hardt & Mishchenko 1931, Wilford 1940, Schwardt 1941), avoiding the use of organic manures at times when the flies are on the wing and the larvae are feeding (Rekach 1932, Harukawa 1933a, Reid 1936, Wilford 1940), and maintaining a dry tilth to discourage the egg-laying flies (Hawley 1922, Rekach 1932).

Experiments on the control of C. cilicrura *in South Worcestershire.*—In preliminary experiments on the control of *C. cilicrura* under field conditions the value of naphthalene as a repellent against the egg-laying flies was tested. An 8-acre bean field was divided into two approximately equal plots, one of which was dusted with naphthalene at the rate of half a cwt. per acre immediately after the seed bed was prepared and again immediately after the ground was harrowed after seeding. The other plot was left untreated, but because the two plots were adjoining it is probable that the control plot was also affected to some extent by the dressings of naphthalene. Counts were made on the treated and untreated plots as soon as the seedlings appeared above ground. Forty counts on areas of 2 sq. yds. (6 ft. lengths of 3 adjacent bean rows) on each plot gave an average of 61 per cent. healthy plants on the treated plot and 53 per cent. healthy plants on the untreated plot. Differences between the two plots were not significant.

On a nearby bean field on the same farm an experiment on the control of bean seed fly with DDT emulsion was carried out. It was intended that, immediately after the preparation of the seed bed, strips of straw or peat 6 ft. wide and 16 ft. apart should be spread across the field (11 acres in extent) and sprayed with a DDT emulsion. Subsequently the strips were to be sprayed with water to keep them moist and attractive to the flies. Because straw would interfere with marking the ground for planting and peat was unobtainable the farmer used straw compost, and after DDT had been sprayed on the compost the weather became showery and both strips and bare soil were kept moist by rain. When the beans showed above ground 40 areas of 2 sq. yds. were examined and the counts showed that an average of 39 per cent. of the plants were healthy. The control plot (on the nearby naphthalene experiment) had 53 per cent. of the plants healthy. The failure of this experiment with DDT does not preclude the possibility of obtaining satisfactory control of the bean seed fly with this substance. Organic manures such as straw compost are considered to be highly attractive to *C. cilicrura*, and during the showery weather that followed its use flies attracted to the field had no need to seek moisture on the treated strips. Wet weather and the nature of the straw compost prevented a satisfactory examination of the soil surface and consequently no information was obtained on the effect of DDT on the flies.

Suggestions for control of C. cilicrura.—This study of *C. cilicrura* suggests that some modification in the timing of operations concerned with planting beans would be likely to lessen attack on the seedlings. The preparation of the seed beds in March should reduce their attraction for the egg-laying flies on the wing about mid-April. Marking the rows should be carried out so as to disturb only dry surface tilth, and harrowing should be delayed until the beans are through the ground or should be followed immediately by the use of a repellent such as naphthalene.

A modification of the calomel treatment against cabbage root fly should be effective in protecting transplanted spring cabbages from attack by the autumn generation of *C. cilicrura*. The roots should be dipped in a paste of calomel dust and water before planting. Dusting with calomel round the collars of newly set plants is not likely to be effective since, unlike cabbage root fly, *C. cilicrura* does not necessarily lay eggs in the proximity of its food-plants and the soil may already be infested at the time the plants are set out.

Acknowledgements.

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Summary.

Field observations on *C. cilicrura* and *C. trichodactyla* have shown that in Britain the larvae feed on beans, peas, cabbages, kale, cauliflowers, leeks, onions, cucumbers and lettuce. The importance of *C. cilicrura* as a pest of beans in spring was well known ; its attack on cucumbers was suspected but had not been verified ; but the infestation of leeks and brassicas in summer and spring cabbages in autumn had not hitherto been recorded in Britain. An association of larvae of *C. cilicrura* with rotting vegetation has not so far been established.

Descriptions of adult and larval stages are given. Diagrams are given to illustrate specific differences between larvae of *C. cilicrura* and those of the onion fly, *Hylemyia antiqua*.

Field observations have shown that *C. cilicrura* is on the wing from March to October and there is some evidence of slight activity during the winter months. There are 3–4 generations a year, and peak periods of fly activity occur during mid-April to mid-May, late June to early July, and in fine weather throughout the period August to October.

Larvae have been taken throughout the period May to December. Peak periods of larval activity occur from mid-May to early June on beans, peas, cucumbers and cruciferous crops, during July on leeks and cruciferous crops, and during September to November on spring cabbages.

The close relationship between cultural practice in south Worcestershire and the occurrence of high populations of *C. cilicrura* is discussed.

A summary of control measures devised by other investigators is given. Modifications of cultural practice that would be likely to reduce attack by *C. cilicrura* are suggested. Results of preliminary trials with naphthalene and DDT emulsion are given.

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EXPERIMENTS ON THE PHYSIOLOGICAL ACTION OF CONTACT INSECTICIDES.

By D. Dresden & B. J. Krijgsman.

Laboratory of Comparative Physiology, University of Utrecht.

Toxicity of DDT, Hexachlorocyclohexane and Rotenone to Insects and other Animals.

Concerning the well-known powerful action of modern contact insecticides the question arises whether these substances exert a specific action on the tissues of insects. It seems quite possible that they possess a general internal toxicity to various groups of animals, but that the insect differs from other animals in that the cuticle is sufficiently permeable to allow the absorption of toxic quantities when these substances are applied to the skin.

In order to solve this problem the action of these insecticides both after application to the skin and after injection must be compared. Such experiments have already been done by some investigators for DDT and Gammexane (pure γ-isomer of hexachlorocyclohexane). Their results are summarised in Table I.

Table I.

LD50 in mg./Kg.

Animal	DDT		Gammexane	
	on the skin	intravenous or intra-abdominally	on the skin	intravenous or intra-abdominally
Rat	3,000	40–50	500	50*
Rabbit	300–2,820	35–50	300	75*
Guinea-pig	1,000	—	—	—
Cat	—	25–40	—	—
Dog	—	60–75	—	—
Periplaneta americana ...	10–30	5–18	4–7·5	3–7·5
Musca domestica ...	7–21	—	0·4–1·2	—
Calliphora sp.	9–28	—	0·6–0·7	—
Aëdes aegypti	5–30	—	—	—

* The LD50 of these animals was estimated after subcutaneous injection. Intravenous injection would, of course, give lower figures.

We have made similar experiments, injecting several hundreds of insects and *Rana* intra-abdominally with water-oil-emulsions of DDT, Gammexane and rotenone. Injection of DDT produced an LD50 of 20 mg./Kg. for *Periplaneta americana*, L., an LD50 of 60 mg./Kg. for *Carausius morosus*, F., and an LD50 of 12 mg./Kg. for *Rana esculenta*. Injection of Gammexane gave an LD50 of 17 mg./Kg. for *Periplaneta* and an LD50 <50 mg./Kg. for *Rana*. With rotenone an LD50 of 6–15 mg./Kg. for *P. americana* and an LD50 of 7–10 mg./Kg. for larvae of *Bombyx mori*, L. (5th instar) was obtained.

Our own experimental results and the data given by other authors are summed up in Table II.

TABLE II.

LD50 in mg./Kg.

Substance	Vertebrates		Insects	
	on the skin	intra-abdominally or intravenous	on the skin	intra-abdominally
DDT	300–3,000	12–75	5–30	5–60
Gammexane	300–500	<50–<75	0·4–7·5	3–17
Rotenone	very high	0·35–2	30 (approx.)	6–15
Strychnine	—	0·2–10	—	500

From this Table some conclusions may be drawn :—

1. A comparison of the LD50 of vertebrates and insects shows the toxicity of DDT to be of the same order. Concerning the toxicity to physiological processes in living tissues, DDT appears to be a general poison and not a specific insecticide. The same conclusion is valid for Gammexane and rotenone, the latter being perhaps a little more toxic to vertebrate animals.

2. When applied to the skin of insects DDT, Gammexane and rotenone display about the same activity as after injection. So the insect cuticle does not protect the animal against these substances. It is not a barrier but permits a quick and complete penetration. Hence the apparent specific toxicity of these poisons to insects.

3. When applied to the skin of vertebrates the toxicity of DDT, Gammexane and rotenone is much lower than after injection. This demonstrates the low permeability of the skin of vertebrate animals to these poisons, so that they are protected by a skin " barrier ".

4. Up to now no poisons have been discovered that possess a specific toxicity to the physiological processes of insects. On the other hand our experiments demonstrate strychnine being, so to speak, a specific " mammalicide ".

Research on new contact insecticides must therefore be guided, given a sufficient toxicity, by their power of penetration through the chitinous cuticle. A high solubility in lipoids is necessary and a specific adsorption by chitin is favourable. The fact that Gammexane is often more toxic to insects than DDT, when applied to the skin (according to our experiments the toxicity of Gammexane as a contact poison for *Calandra granaria* is about 60 times higher) is obviously also based on a different rate of penetration.

Insecticidal Properties of the Isomers of Hexachlorocyclohexane.

Slade (1945) and Jenkins (1945) proved the α-, β- and δ-isomers of hexachlorocyclohexane were not toxic in moderate concentrations to insects and mammals when applied as a contact poison. Under these circumstances only the γ-isomer (Gammexane) displayed a powerful insecticidal action. Our own experiments, to be published elsewhere, fully confirm Slade's results.

The question arises whether the difference in activity of these isomers is caused by a difference in rate of penetration through the cuticle or by a difference in real internal toxicity to tissue processes. It is quite possible that all isomers have a similar range of internal toxicity but that only the γ-isomer permeates quickly enough through the cuticle to exert a toxic influence after skin-application.

In order to settle this question injection experiments were carried out with the pure isomers *(water-oil-emulsions) with *Periplaneta americana.* The LD50 for the γ-isomer appears to be 17 mg./Kg. The toxicity of the other isomers is much lower, as a dose of 85 mg./Kg. did not produce any effect. It was impossible to make emulsions with a higher content of these isomers, so the real LD50 could not be estimated.

The conclusion is drawn that the α-, β- and δ-isomers at best have a low rate of toxicity to insect tissues. Thus the high insecticidal value of the γ-isomer cannot be the consequence of a quicker penetration, but must be based on the special stereo-configuration of its molecule which allows a specific attack on unknown physiological processes.

The Physiological Relation between Gammexane and Meta-inositol.

The γ-isomer of hexachlorocyclohexane (Gammexane) possesses a molecular structure very similar to that of meta-inositol. This occasioned Slade (1945) to postulate the hypothesis of competition between Gammexane and meta-inositol in cell metabolism. Buston, Jacobs & Goldstein (1946) and Kirkwood & Phillips (1946) brought some experimental evidence to support this hypothesis by showing that Gammexane inhibits the growth of yeast. The inhibitive action of Gammexane could be abolished on adding inositol to the medium.

Inositol is a necessary growth factor in the case of yeast, but for animals, especially insects, this has never been verified. We only know that an inositol-free diet causes alopecia in rodents, without being a lethal factor. Still it might be possible that meta-inositol is indispensable to insects and could be ousted from vital structures in cell metabolism by Gammexane. If this be right, then we may expect that the simultaneous application of both substances to insects lowers the toxicity of Gammexane, in other words the LD50 of Gammexane must increase.

Such experiments were carried out with *Periplaneta americana.* A large number of these insects was injected with the LD50 of Gammexane (17 mg./Kg., water-oil-emulsion) ; a second group was given this amount of Gammexane and at the same time as much meta-inositol ; a third group was given only this quantity of meta-inositol. No difference was found between the first two groups, the mortality being 50 per cent. in both cases. The cockroaches injected with inositol showed only a low mortality. The experiment was repeated by injecting 17 mg./Kg. Gammexane and 34 mg./Kg. meta-inositol simultaneously. In this case also there was no protecting influence by inositol, mortality being 50 per cent. again.

The results of these experiments make it improbable that Slade's hypothesis is true.

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NOTES ON THE GREEN LEAFHOPPER, *EMPOASCA LYBICA*, BERG. (HOM. JASSID.) IN PALESTINE.

By H. Z. Klein, M.Sc.A., D.Sc. (Jerusalem).

Jewish Agency for Palestine, Agricultural Research Station, Rehovot.

The Green Leafhopper, *Empoasca lybica*, Berg. (=*E. benedettoi*, Paoli, *Chlorita signata*, Haupt) has so far been found in Spain, Tripolitania, Cyrenaica, Egypt, Sudan, Eritrea, Somalia, Tanganyika, Uganda and Palestine.

From 1942 to 1945 research work was carried out in Rehovot on the life-history of this Jassid on egg-plants under outdoor conditions. The plants were grown in flower-pots and covered with glass bell-jars, the lower ends of which were inserted into the soil and the upper ends covered with thin cloth.

Life-cycle of *E. lybica.*

Oviposition.—The leafhoppers copulate in the hot season one or two days after the last moult. Copulation takes from half to one hour, but sometimes lasts for several hours. Parthenogenetic reproduction was never observed. One copulation is sufficient to ensure repeated ovipositions and females isolated after their first mating lay as many eggs as those mated several times.

The pre-oviposition period varies from two to three days in summer to three weeks or more in winter. The eggs are laid in the leaf tissue, especially in the main veins of the under side of the leaf, and in the case of the egg-plant no discoloration marks the place of oviposition. The average number of larvae that hatched from eggs laid by 65 females was 16, the maximum number per female being 81. The natural mortality of the eggs is small, as they are laid in well-protected places. The maximum oviposition was seven eggs per day with a daily average of one to three eggs per female.

Egg development in summer lasts from 5 to 15 days, in spring from 9 to 22 and in winter from 20 to 44.

Larval development.—The head of the hatching larva appears first above the leaf surface with its red eyes conspicuous. After an interval of a quarter of an hour or more the body emerges hastily from the leaf tissue, the whole process of hatching generally taking nearly half an hour. The legs are freed (pair after pair, starting with the first pair) by bending movements of the body. A few seconds after emergence, the young larva becomes active and migrates until it begins to feed.

The larval development lasts, according to the season, from 8 to 50 days, during which period it moults five times. The first stage is the shortest, and its duration is 15 per cent. of the whole period of larval development ; the second, third and fourth stages are equal in duration, each lasting about 18 per cent. whilst the last stage is the longest and takes 30 per cent.

The number of offspring from each pair of parents was in spring, 7–13 ; in summer, 2–17 ; in autumn, 2–57 ; and in winter, 4–81 ~~days.~~

Number of annual generations.—The total development, from egg to adult, varied from 15 days in summer to 72 in winter.

Table I shows the sequence of the generations for two years.

TABLE I.

Appearance of the adults of the various generations in 1940–1944.

No. of generation	Date of appearance of adults of each generation	No. of days between oviposition of one generation to that of the next
	5.x.42	36
1	10.xi.42	55
2	4.i.43	84
3	29.iii.43	76
4	13.vi.43	35
5	18.vii.43	23
6	10.viii.43	20
7	30.viii.43	21
8	20.ix.43	29
9	19.x.43	40
10	28.xi.43	144
11	20.iv.44	64
12	23.vi.44	30
13	23.vii.44	19
14	11.viii.44	26
15	6.ix.44	30
16	6.x.44	

In two successive years the Green Leafhopper gave rise to 16 complete generations, i.e., an average of eight annual generations. The first offspring of the tenth generation matured at the end of November 1943, while the last offspring of the same generation only reached maturity in April 1944. The leafhoppers that matured in late November actually laid eggs that hatched, but most of the larvae died during the cold season ; it was the later hatchings that gave rise to the new generation in the spring.

There is no marked difference in longevity between the males and the females, and isolated females lived, at all seasons, as long as copulated females. In summer the adults lived, on the average, about two weeks and in winter up to seven weeks ; some of the leafhoppers, maturing in autumn, over-winter. The sex ratio was about equal throughout the year.

Influence of Climate on Development.

Pre-oviposition period.—The influence of climatic factors on the length of the pre-oviposition period during the year can be seen in Table II.

TABLE II.

Duration of the pre-oviposition period in days.

Season	Duration of the period		Temperature in °C.	R.H. in per cent.
	Minimum	Maximum		
Spring	4	5	23·5	70
Summer	2	3	25	67
Autumn	6	13	19	64
Winter	21	23	14–15	73

In Rehovot, the average air humidity during the breedings was relatively uniform between 64–73 per cent., and the influence of the air humidity, therefore, is not conspicuous. No eggs at all were laid on khamsin-days (dry east wind), when air humidity is very low and temperature high.

Temperature has a very definite influence on the pre-oviposition period, the duration of which is prolonged with the decrease of temperature, from two to three days in summer up to three weeks in winter.

Egg development.—The length of the stage at different times of the year is shown in Table III. Air humidity has no influence, as the eggs are developing in the tissues of the leaf and are therefore not affected ; the eggs depend on the moisture of the leaves, and die immediately the latter wither.

TABLE III.

Duration of development in days.

Month of start of development	Length of egg stage	Length of larval stage	Total development
February	20–27		62–65
March	27	27–33	
May	9–15	12–17	26–32
June	10–14	11–16	25–27
July	7–10	9–16	18–24
August	8	8–11	16–18
September	5–13	9–14	15–26
October	7–15	10–16	18–34
November	10–22	12–33	27–52
December	19–44	26–50	67–52

The influence of temperature on the duration of the egg-stage can be seen in Table IV. In the hottest season the larva hatches from the egg after five days, while at a low temperature it may take as long as seven weeks. Egg mortality is very high during the cold winter months (January and February), as during this season the leaves of the egg-plant wither to a large extent. Also, in August, the hottest month, when the average monthly temperature approaches 27°C., there is apparently a great mortality of the eggs or young larvae.

TABLE IV.

Temperature and duration of the development in days.

Temperature in °C.	Egg		Larva		Total development	
	Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
13	19	49	—	—	—	—
14	—	—	26	50	55	72
15	20	27	—	—	—	—
16	—	—	—	—	45	60
17	—	—	27	33	—	—
18	—	—	—	—	35	43
19	10	22	16	33	—	—
20	—	—	—	—	27	34
21	9	15	14	17	—	—
22	—	—	12	19	26	32
23	—	—	11	16	23	29
24	7	15	—	—	15	27
25	—	—	10	14	18	26
26	5	13	9	15	15	18
27	—	—	8	16	—	—

Larval development.—Table III shows also the details of larval development in the various months. Air humidity had again no influence but, on khamsin-days, when it was very low, many larvae died and especially those of the first stage. The duration of larval development depends, mainly, on temperature (Table IV) ; at the hottest season the larva can reach maturity in eight days, but needs five to seven weeks for its development in winter.

Larvae, particularly very young larvae, died in large numbers in December and January, when the monthly average temperature was under 14°C. The temperature in these months reached a minimum of 7°C., and at this temperature the larvae froze to death.

In August, at a monthly average of almost 27°C., the larvae died because of the withering of the leaves due to the dry heat.

Total development.—For the details of the total development throughout the year see Table III.

Table IV shows the influence of the temperature on the total development. The shortest development of *E. lybica* is in 15 days at the height of the summer. In winter almost two and a half months are needed for the development of one generation.

The threshold of all stages, as well as the thermal constant, was calculated from the breeding results.

	Pre-oviposition period	Egg development	Larval development	Total development
Threshold of development ...	11·0	10·0	12·0	11·0°C.
Thermal constant	43	125	150	280 day-degrees

In all stages the threshold of development is between 10–12°C. The thermal constant needed for the development is 280 day-degrees.

Activity of the adults.—The activity of the adults in the field is greater in the summer months, and especially during the hottest time of the day. When one touches the plant at these hours the adults rise quickly from the leaves and fly to another plant.

The leafhoppers are found in the summer months mainly on the upper leaves, either because of their tenderness or because the temperature is not so high as it is nearer the ground. The adults and larvae at all stages feed on the lower side of the leaves, thus securing protection from the sun. It is difficult to catch the adults at the hot time of day because of their great activity.

Activity experiments were made according to Bodenheimer's method to determine the temperature limits of the Green Leafhopper. A large number of adults and larvae of various ages were heated slowly in test-tubes in a sand-bath until they died. Both adults and larvae quickly rose to the vicinity of the stoppers, where the temperature was measured near them. The larvae, and especially the adults, were very active and alert in the test-tubes during the experiments. While jumping some of them reached the bottom of the test-tubes, where the temperature, being, of course, much higher than near the stoppers, killed them immediately. For these experiments, which commenced at a temperature of 25°C., a hundred insects were used. Table V shows the behaviour of the insects at the various temperatures, according to stage of development.

TABLE V.

Degree of activity	Adults	Large larvae	Medium larvae
Upper limit of normal activity	32°C.	32°C.	30°C.
Increased activity	33–37	33–36	32–35
Maximum of excitement	38–44	37–43	36–39
Immediate heat-death	46–47	45–46	41–43

According to these results, *E. lybica* is more sensitive to heat the younger it is. The medium larva dies at a temperature of 5°C. lower than the adult. This explains why young larvae in the field in the hottest weather lack mobility and are found on the under side of the leaves. The adults, on the other hand, jump a long distance at such times, especially when the leaves, on which they sit, are touched.

In the winter months hardly any Green Leafhoppers are found on cultivated plants, and on wild herbs they are very scarce in the cold season.

In breeding experiments under outdoor conditions it was found that all movement of adults ceases when the temperature drops below 14°C., and oviposition commences only when the daily average temperature rises above 15°C. During khamsin-days no eggs are laid.

Host Relations.

Vine, cotton, sugar-cane, sorghum, sunflower, lucerne, etc., are recorded from various countries as suffering damage from this pest. In Palestine it is particularly harmful to egg-plant, tomato, pepper and potato and also occurs on beets, radishes, sweet potato and various pulses and species of Cucurbitaceae. It is not yet known if the abundant Green Leafhoppers on vine and castor plant belong to the species under discussion. (Bodenheimer mentions the species *Chlorita flavescens*, F., on the castor plant.)

The Green Leafhopper undoubtedly infests various wild herbs in Palestine, but few observations have been made ; it is possible that they are important winter hosts of the pest. It should be mentioned that the nightshade is an important host plant.

The symptoms of damage on egg-plant are, yellow stains on the leaves which later turn brown, and curl at the edges. This phenomenon is caused by the feeding of the adults and larvae on the underside of the leaves which are attacked chiefly near the edges and in the middle near the main vein.

The leafhoppers appear in masses in summer and autumn when they cause great damage. With the beginning of the rainy season their number greatly decreases, and the damage becomes small and economically unimportant.

Although large egg-plants, planted at the beginning of the summer (from May to early June), do not suffer much, serious damage is sometimes caused to young plants in the middle of summer (mid-June to mid-August). In such cases the flowering and development of the fruit is delayed to such an extent that it is not worthwhile to continue the cultivation of the plants.

In tomato nurseries light streaks, caused by the feeding of the hoppers, appear on the stems of the plants and they may spread over the entire stem. Such affected stems become brittle and are easily broken.

It is not yet known whether the Green Leafhopper transmits virus diseases in Palestine but the curling of the leaves, stains resembling those of mosaic disease, and shedding of flowers and fruits immediately after formation, are suspicious symptoms.

Control.

In 1928 successful control experiments were carried out by Bodenheimer and Klein with a Veratrin compound, an alkaloid extracted from the roots of *Veratrum album*. This compound disappeared from the market, and in 1938–42, therefore, experiments with compounds of rotenone, nicotine, whale-oil soap, Bordeaux mixture and sulphur were made. These experiments, however, did not give entirely satisfactory results, even when the treatment was repeated several times during the season.

Summary.

In the coastal area of Palestine, *Empoasca lybica* has eight annual generations. The average number of offspring per female is 16, 81 being the maximum. The adults live in summer up to seven weeks whilst those of the autumn-generation overwinter and live up to five months. The pre-oviposition period, the development of the egg and larva and the total development last from 2 to 23, 5 to 49, 8 to 50 and 15 to 72 days, respectively, according to the season of the year. The threshold of development is between 10 and 12°C. in the various stages of development. The duration of development depends mainly on the temperature. Air humidity has no influence on the duration of development so long as the leaves are sufficiently succulent. Activity of the adults ceases below 14°C. and a temperature above 46°C. results in immediate death. During khamsin-days no oviposition takes place, and many larvae die, especially those of the first stage. The most important local hosts are Solanaceous plants. The characteristic damage is the appearance of yellowish stains on the leaves and leaf-curling ; the plants are delayed in development, and shed their flowers and small fruits. Young plants wither when heavily attacked. Damage is most severe in young plants in mid-summer. So far no satisfactory control measures have been found in Palestine.

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PRELIMINARY TESTS OF DDT EMULSION CONCENTRATES.

By B. M. Jones, B.Sc., F.R.E.S.

Lieutenant (Sp.) R.N.V.R. Medical Department of the Navy.

DDT emulsion concentrates, designed for dilution with water for spraying, are of two types; in both, the solvent used for the DDT is one in which it is highly soluble. One, the "soluble-oil" concentrate, consists of a solution of DDT and emulsifier in the solvent and forms a homogeneous emulsion when dispersed in water. The other, the "mayonnaise-cream" concentrate, is itself a concentrated emulsion, since the solution of DDT is already dispersed in water with the aid of an emulsifying agent. At the time when these concentrates were first introduced for field trials, a kerosene-oil base was universally considered, in the Services, to be the most economic and efficient carrier-medium for DDT in the control of mosquitos and flies, and statements had been made that emulsified solutions of DDT were not producing the results required in the field. It was felt, however, that there was no reason why DDT should not be at least as effective in an emulsion as in a homogeneous oil base, provided that the emulsion concentrate was shown to satisfy the requirements of a standard test for physical stability. Advantages of the emulsions are that the concentrate can be diluted with water in the field, thereby easing transport, that the emulsions are less toxic to operators than kerosene solutions of DDT, which sometimes cause kerosene dermatitis, and that their inflammability is less.

The following four emulsion concentrates, which were received by the R.N. School of Tropical Hygiene, Colombo, in 1945, are chosen to illustrate the kind of tests to which samples were submitted before being tried on a field scale. The DDT in all of them was of technical grade (60·5 per cent. pp′isomer).

Emulsion Concentrates.

Emulsion Concentrates	No. 1 "Soluble-oil" type	Emulsion Concentrates	No. 2 "Mayonnaise-cream" type
	Per cent.		Per cent.
DDT Technical	23·2	DDT Technical	30·0
Solvent naphtha	38·2	Solvent naphtha	40·01
Teepol X	9·0	Casein	1·45
Liquid rosin	15·0	Phenol	0·64
Cresylic acid	9·0	Agral W.E.M.	0·50
Caustic soda	1·8	Caustic potash	0·22
Ethyl alcohol	2·0	Ammonia (0·88)	0·38
Water	1·8	Water	26·80

Emulsion Concentrates	No. 3 "Soluble-oil" type	No. 4 "Soluble-oil" type
	Per cent.	Per cent.
DDT Technical	25·0	25·0
Toluene	33·35	33·5
Turpentine	33·35	25·6
Ground-nut oil	—	7·6
Rectified spirit	4·9	4·9
Dry soap	2·8	2·8
Water	0·6	0·6

Physical Stability Tests.

For practical use, the emulsions formed should be reasonably stable at concentrations of 1 per cent. to 10 per cent. DDT ; extreme stability is not necessary for most purposes. For the purpose of these tests an emulsion was considered satisfactory if at 1 per cent. DDT concentration, no visible separation occurred within 2 hours and if at 24 hours only moderate " creaming " had occurred (both layers " creaming "), with no separation of the oil phase. Emulsions containing 1 per cent. DDT were prepared by adding 5 ml. concentrate slowly, with shaking, to the required quantity of fresh water in a 500 ml. Erlenmeyer flask. The contents were shaken vigorously for one minute after the addition of the concentrate, and 100 ml. was then transferred to a 100 ml. measuring cylinder for observation.

TABLE I.
Behaviour of 1 per cent. DDT emulsions in fresh water.

Time	No. 1 Soluble oil	No. 2 Mayonnaise	No. 3 Toluene/Turpentine	No. 4 Toluene/Turpentine and Ground-nut oil
15 min.	Stable ...	Stable	Settling into two layers ...	Same as (3)
30 min.	Stable ...	Stable	Two distinct layers ...	Same as (3)
1 hr.	Stable ...	Stable	Oil phase appearing as visible globules	Same as (3)
2 hrs.	Stable ...	Stable	Visible oil globules increasing	Same as (3)
24 hrs.	Stable	Stable (formation of a thin clear surface layer)	Oil phase surface layer evaporating — DDT recrystallising	Oil phase as a surface layer of large globules

Results show that No. 3 and No. 4 " soluble-oils " are out of specification. Tests were duplicated six times.

Tests repeated with dilutions to 5 per cent. DDT concentration gave identical results.

Practical tests with 5 per cent. DDT emulsions prepared from No. 3 " soluble-oil."

A quantity of 5 per cent. DDT emulsion was prepared according to the standard method of adding 1 part concentrate to 1 part water and then adding the required volume (in this case 3 parts) of water with vigorous stirring, and 10 litres of the prepared emulsion was transferred to a Four Oaks " Ross " Knapsack container. Observations of the behaviour of the emulsion were made on samples taken by means of a " dip " tube from the container while it was worn by an operator carrying out routine spraying.

TABLE II.
Physical stability of 5 per cent. DDT emulsion subjected to normal agitation in a Knapsack Sprayer while being used for routine spraying.

Time	Behaviour
15 min.	Signs of formation of two layers
30 min.	Two layers forming
40 min.	Two layers but no distinct demarcation
1 hr.	Two distinct layers. Oil globules beginning to form on the surface of the solution

TABLE III.

Physical stability of 5 per cent. DDT emulsion subjected to special shaking of the Knapsack container, at 5 min. intervals, while being used for routine spraying.

Time	Behaviour
15 min.	No visible separation into two layers
30 min.	Two layers forming
1 hr.	Two layers (both creaming)
2 hrs.	Two layers but no distinct demarcation Oil globules on the surface of the solution

The behaviour of this 5 per cent. DDT emulsion, prepared from No. 3 toluene/turpentine " soluble-oil ", when observed in a knapsack sprayer under normal conditions, was similar to that shown in the laboratory tests. Stability was slightly improved when the container was shaken at 5 min. intervals. Moreover 100 ml. samples of the emitted spray were taken after the emulsion had been allowed to stand for 30 min. in the knapsack container. The range was represented by a series of samples, with the first samples taken being more or less water and those at the end of the range having a high percentage of the oil-phase solvent containing the DDT.

Behaviour of concentrates when dispersed in salt water.

For the control of mosquito vectors of malaria that breed in water with a high salt content, *e.g.*, *Anopheles sundaicus* (Rodenw.), it is an additional and important property if the concentrate will disperse in brackish or sea water to form a homogeneous emulsion. The data in Table IV were obtained by using salt water from a lagoon in which *Anopheles subpictus* (Grassi) was breeding prolifically. The salinity of the lagoon water was 4·5 per cent. (expressed as NaCl) and although the test was a rigorous one, identical results were obtained with sea water (salinity 3·5 per cent.) and within a descending range to water with a salinity of 0·5 per cent.

TABLE IV.

Physical stability of 1 per cent. DDT emulsion in salt water (salinity 4·5 per cent. expressed as NaCl).

Time	No. 1 Soluble oil	No. 2 Mayonnaise cream	No. 3 and No. 4 Soluble oils
30 sec.	Oil phase separating as visible globules rising to surface	Stable	Oil phase separates immediately into large globules which rise to the surface
1 min.	Oil phase completely separated as a surface layer	Stable	Oil phase completely separated as a surface layer
1 hr.	—	Stable	—
2 hrs.	—	Stable	—
16 hrs.	—	Emulsion globules concentrated as a bottom layer; emulsion could be reformed but it settled out completely in 15 min.	—

The results show that only the " mayonnaise " concentrate formed a homogeneous emulsion when dispersed in salt water.

Persistent toxic effect on mosquitos of residual films of two emulsions.

Emulsions were prepared from No. 1 (soluble oil) and No. 2 (mayonnaise) concentrates, and their potency, when applied as a surface film, was measured by (*a*) the dosage-biological activity relationship, (*b*) the response and susceptibility of the test insects under standard conditions indicated by the time required to give a 100 per cent. kill. The mosquitos used were *Armigeres obturbans* (Wlk.) and *Culex gelidus* (Theo.). It was the aim to conduct the experiments under as natural conditions as possible without detracting from the need for standardisation. Testing rooms were improvised by constructing false ceilings of mosquito netting in three outbuildings, which were in a state of good repair, with plaster walls, stone floor and a wooden door to each. Previously these rooms were natural resting places for mosquitos and provided a source of supply as a catching station. Two testing rooms were of equal size, each with a treatable surface of 132 sq. ft. Another similar room served a dual purpose ; it was used to breed the *Armigeres obturbans* from the pupae and became a control test for the experiment. Adults of *Culex gelidus* were collected from nearby natural resting places.

The concentrates were diluted to emulsions containing 5 per cent. DDT by mixing 1 part concentrate with 1 part water and then adding the required volume of water. An " Aeraspray " pressure gun was used to apply the emulsion as a film on the plaster walls and door surfaces. The spray was applied evenly ; sufficient to wet the surface and avoid streaming. Since the sprayed area and quantity of emulsion used were known, it was possible to calculate the approximate deposit of DDT per sq. ft. A more accurate estimate of determining the deposits of DDT was made by placing weighed glass slides in the path of the spray. The glass sides were immediately replaced in stoppered tubes and reweighed. From the weight of deposit on the glass slides, the deposit of DDT in mg./sq. ft. was calculated.

On the second day after treatment 30–40 mosquitos were introduced each day and as far as possible an equal number of the two sexes per batch. A clean white sheet was laid over the floor surface to facilitate the observations and count of " knock down ". The insects were collected at 15-minute intervals and observations

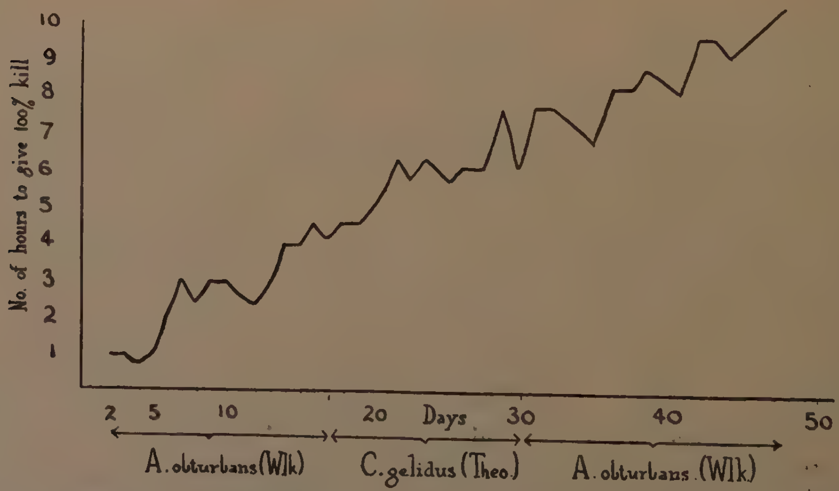


Fig. 1.—Graph showing toxic effect of residual film of No. 1 " Soluble-oil " emulsion on *Armigeres obturbans* and *Culex gelidus*.

were made to determine whether there was a recovery rate. During the tests over the period recorded not one " knock down " insect recovered. Therefore " knock down " is defined as kill in the graph results. Occasionally escape by the test insects accounted for the recovery of less than the total number introduced, but the number lost, except on one occasion, never exceeded two or three.

Treated surface area of testing room	132 sq. ft.
Strength of DDT emulsion	5 per cent.
Rate of concentration of DDT deposit	186 mg./sq.ft.
Mean temperature	80°F.
Mean relative humidity	78 per cent.

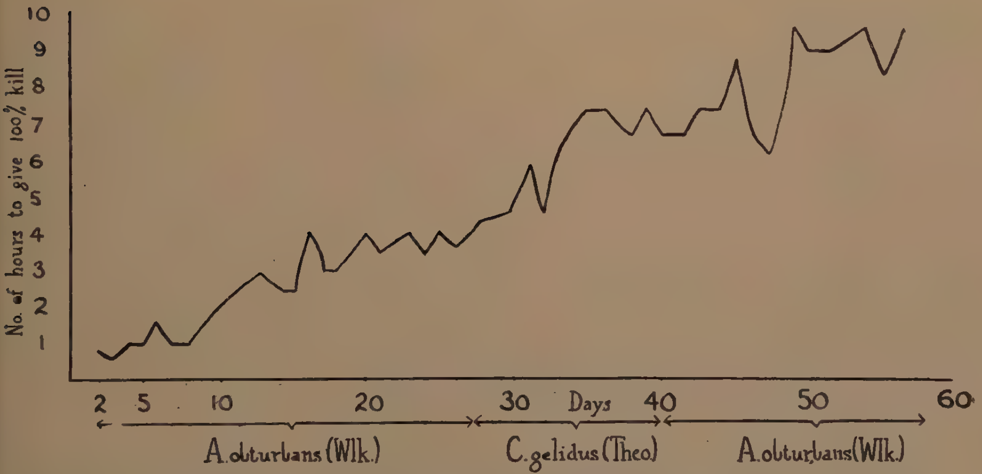


Fig. 2.—Graph showing toxic effect of residual film of No. 2 " mayonnaise " emulsion on *Armigeres obturbans* and *Culex gelidus*.

Treated surface area of testing room	132 sq. ft.
Strength of DDT emulsion	5 per cent.
Rate of concentration of DDT deposit	206 mg./sq. ft.
Mean temperature	80°F.
Mean relative humidity	78 per cent.

The de-activation of the DDT deposit is shown by an increase in the time required to give 100 per cent. " kill " which is inversely proportional to the persistent toxicity of the film.

It was not convenient to observe and make counts after a 10-hour period in one day. But the results over a protracted period of 50–55 days showed that the persistent toxicity of a residual film resulting from the spray application of a 5 per cent. emulsion compares favourably with that of a 5 per cent. kerosene solution.

Summary.

A method for testing the physical stability of emulsions is described.

Since the persistent toxicity of residual films is so important in the control of mosquitos and flies, this biological activity of stable DDT emulsions was measured by the response and susceptibility of *Armigeres obturbans* and *Culex gelidus* over a protracted period of time.

These discriminating tests were carried out before emulsion concentrates were tried out in the field.

FIELD EXPERIMENTS WITH DDT AND BENZENE HEXACHLORIDE AGAINST TSETSE (*GLOSSINA PALPALIS*).

By C. B. Symes, A. B. Hadaway, F. Barlow and W. Galley.
Colonial Insecticide Research.

(Plate XI.)

D G.

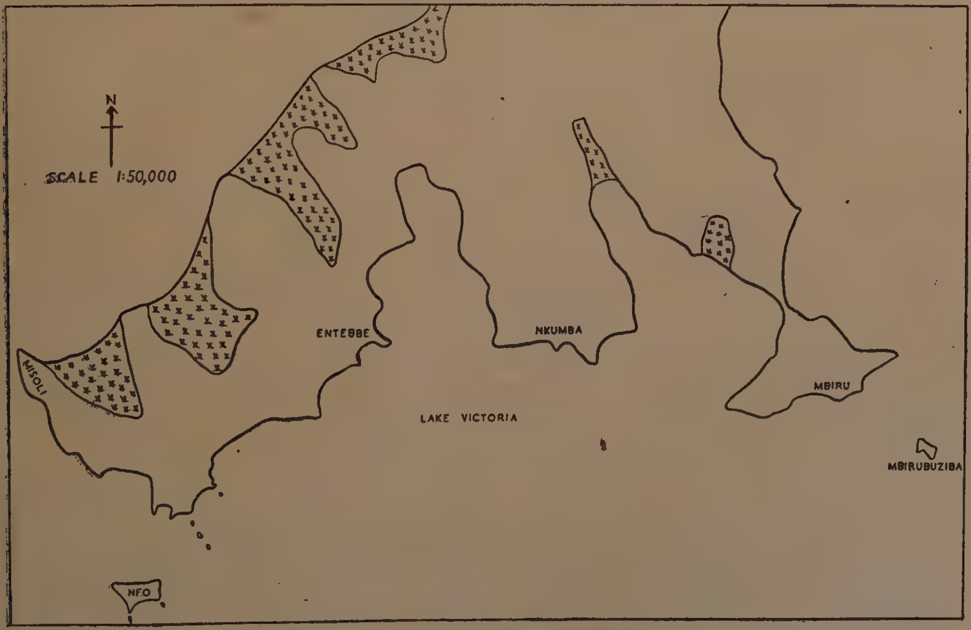
Applications to Vegetation by Hand Spraying.

Field experiments have been conducted in Uganda to investigate the use of the insecticides DDT and Benzene Hexachloride for the control of tsetse. Since the Tsetse Research Department of Shinyanga has developed a line of investigation into the possibility of using cattle sprayed with DDT as poison bait for tsetses, it was considered that the alternative method of attack, i.e., treatment of vegetation, should be tried. The preliminary work has been against *Glossina palpalis*, R.-D. Results of experiments against this species should provide a basis for work against other species. Experiments have been carried out as far as possible on small islands and headlands on Lake Victoria where the flies exist in isolated communities and where results are not therefore influenced by infiltration of flies from surrounding bush.

In the first experiments the effect of a single application of insecticide to vegetation was observed. In later ones four applications were made in an attempt to maintain a toxic deposit over the maximum pupal period of about 55 days.

Field Experiment 1. Nfo Island. (Map 2.)

Nfo Island lies on Lake Victoria about one mile from the mainland south of the Wireless Station, Old Entebbe (Map 1). It measures 1,000 yards east to west; 600 yards north to south at the widest point. The area is approximately 100 acres.

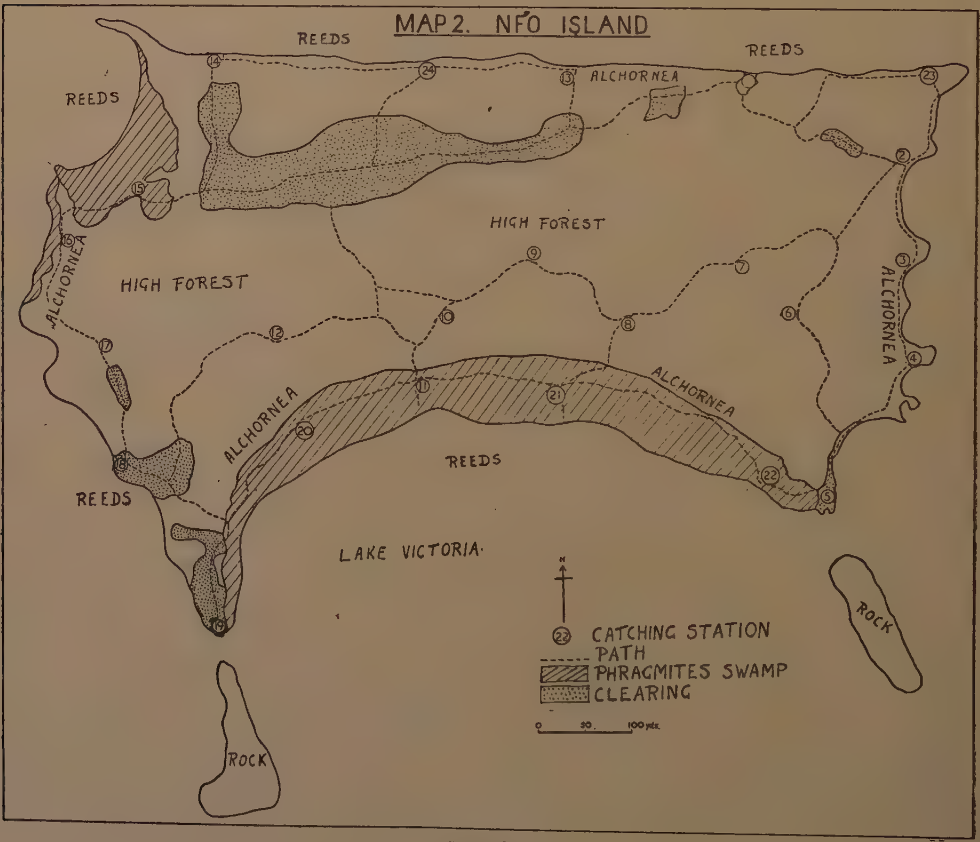


Map 1.

The island is uniformly flat, a few feet only above lake level, except the south-western headland which rises to about 15 feet. The centre of the island has a canopy of high trees, up to 60 feet in height, with a dense undergrowth of plants, shrubs, and creepers. Generally this is bordered by a belt of lower trees (20–30 feet) which reaches to the lake shore on the north coast. Outside this belt on the western and southern sides are swamps with a very dense growth of plants, up to 10 feet in height, traversed by numerous hippo paths. The south-west and north shores are fringed with reeds and water-lilies.

Some small sandy beaches with overhanging rocks and vegetation occur on the coast line; they are most numerous between catching stations 2 to 5, and 18 to 19. These probably provide the chief breeding sites.

One extensive treeless area and several small ones, where the dense vegetation is replaced by short grass on sandy soil, occur on the north and west sides a short distance from the lake edge (Map 2).


MAP 2. NFO ISLAND
REEDS
REEDS
REEDS
ALCHORNEA
HIGH FOREST
HIGH FOREST
ALCHORNEA
ALCHORNEA
ALCHORNEA
ALCHORNEA
REEDS
REEDS
LAKE VICTORIA
ROCK
ROCK
N
㉒ CATCHING STATION
------ PATH
▨ PHRAGMITES SWAMP
░ CLEARING
0 50 100 yds.

Map 2.

Albizzia zygia (Mimosaceae) is predominant in the canopy of trees, whilst *Ficus dawei* (Moraceae) is common along the lake shore. The undergrowth consists mostly of an association of four plants :—*Haemanthus multiflorus* (Amaryllidaceae), a low plant with glabrous leaves up to 12 inches long and 3 inches broad ; *Aframomum* sp. (Zingiberaceae), with large glabrous leaves, 18 inches long, arranged in two rows on a stem up to 15 feet in height ; *Culcasia scandens* (Araceae), a creeper with glabrous elliptic leaves 5 to 10 inches long and 2½ to 4 inches broad; and *Justicia* sp. (Acanthaceae). *Alchornea cordata* (Euphorbiaceae) (Pl. XI, fig. 3) occurs extensively round the periphery of the island, and is the most common plant. It is a shrub growing up to 25 feet in height, bearing many broadly ovate leaves 4–10 inches long and 3–6 inches wide. Another common shrub is *Triumfetta macrophylla* (Tiliaceae) (Pl. XI, fig. 2). It grows to a height of 15 feet and bears very hairy leaves up to 9 inches long by 6 inches broad.

The swamp areas consist of a dense growth of numerous plants, *Phragmites mauritianus* (Gramineae), *Polygonum* sp., *Melanthera brownei* (Compositae), *Mimosa pigra*, and *Sesbania sesban* (Papillionaceae). Papyrus (*Cyperus papyrus*) and Ambatch (*Herminiera elaphroxylon*) occur on the extreme edge of the swamps.

Hippos are often seen off the east and west shores; their tracks are very numerous in the swamps on the south and west. Crocodiles have been observed on the rocks of the south-east. Monitor lizards (*Varanus*) are numerous and occur over most of the island. Cormorants are common on the east coast, but bird life is not plentiful. A few fish eagles are usually to be seen.

Mosquitos are numerous throughout the island. Many, with *Taeniorhynchus africanus*, Theo., predominant, bite vigorously by day. Numbers of lake fly (Chironomids) fluctuate. Dragonflies of various species are abundant over the clearings and less so along the cut paths. Distribution of butterflies is similar. General studies of insect population have not been made.

The island is not occupied and there is no cultivation.

Paths were cut round the island as near the lake shore as possible and across the island in various directions. Sample fly catches (*G. palpalis*) were made along these paths and 24 catching stations were then established at intervals for daily density catches. The latter were based on two boys catching for 15 minutes at each station; flies were released at the end of each catch. The fly population, estimated by marking and recapturing, was 11,700. Fly densities, before and after treatment can be seen in fig. 1.

Density catches were made daily at 16 catching stations set up in a small isolated belt of lake shore bush on the mainland at Misoli some three miles from the island (Map 1). This area remained untreated. Weekly mean number of flies per station is given in fig. 1.

Application of Insecticide 23.xii.45.

Insecticides.—Gammexane (0·65 per cent. gamma) solution in 50 per cent. power kerosene and 50 per cent. cottonseed oil. This was made up by dissolving D.929 at the rate of 8 lb. to 3 gals. of power kerosene heated over a water bath to 40–50°C., and adding the concentrate to a cold mixture of 5 gals. of power kerosene and 8 gals. of cottonseed oil. An appreciable amount of D.929 was insoluble.

Spraying Apparatus.—Four Oaks " Kent " Sprayers, each fitted with rubber hose and metal nose piece bearing two nozzles, and each having a capacity of 1¾ gals. Hand pumped to a pressure of 75 lb. per sq. inch, the container empties in approximately three minutes continuous spraying. Experiments showed that at an average rate of progress 150 sq. yards were sprayed in 80 seconds. On this the area sprayed is estimated, but in this and in the following experiments the estimations of dosage per foot are most probably too good to be true.

Dosage.—Total material used—48 gals. of solution. Total spraying time 87½ minutes (between 9 a.m. and 3 p.m.).

Approximate area covered.—88,450 sq. ft., at the rate of 2·2 qts. per 1,000 sq. ft., or approximately 11 mg. of gamma isomer per sq. ft. For the whole island, dosage was at the rate of 0·5 ounces of gamma isomer per acre.

Area sprayed.—Spraying was confined almost entirely to the periphery of the island at 60 selected fly concentration points (as determined by sampling density catches), along hippo tracks, on rocks and overhanging vegetation. Vegetation was treated to a height of approximately 6 ft.

General Observations.—No repellency to tsetse flies was apparent; they constantly alighted on operators during spraying.

Leaves of *Alchornea cordata* showed no signs of burning; even after three weeks the upper leaf surfaces of the plant were still shiny and "tacky". In general, glabrous leaves showed little burning, but in places whole shoots of plants with soft hairy leaves quickly turned brown and died.

There was a noticeable temporary reduction of dragonflies over the clearings, and of mosquitos in the staff hut.

On the day of spraying there was a hot sun, and cloudless sky, with a strong northerly wind in the afternoon. No meteorological instruments were available on the island, but records taken on the mainland, 2 miles from the island, showed a mean daily maximum temperature for January—80·3°F., a minimum temperature of 63·9°F., and a relative humidity at 8.30 a.m. of 83 per cent. and at 2.30 p.m. of 62 per cent. Rain occurred on 25.xii.45, 0·07 in.; 27.xii.45, 0·04 in.; 28.xii.45, 0·02 in.; 5.i.46, 0·04 in.; and 22.i.46, 0·15 in.

The method used for chemical determination of both DDT and Gammexane has been dehydrohalogenation followed by titration of the liberated chlorine ions with silver nitrate solution. The leaves were first washed with benzene. Using benzene as a stripping solvent, the percentage recovery of known weights of insecticides applied to leaves of mango, *Hibiscus*, avocado pear, *Alchornea cordata* and *Triumfetta macrophylla* was 99–100 per cent.

The test insect, *G. palpalis*, was bred in the laboratory from pupae collected on the lake shore. They were given the opportunity to feed on sheep daily. Flies 7 to 10 days old were exposed to the treated surface individually in 3×1 inch tubes, which were then kept in racks over water in special trays. Mortality counts were made 24 and 48 hours after contact.

Laboratory techniques in this first series of tests were probably not good. Samples of treated leaves were brought into the laboratory at various times after the application of insecticide for chemical estimations of surface deposits. The amounts of insecticide recovered, given in Table I, were low.

TABLE I.

Chemical determination of deposits on leaf surfaces from Nfo Island.

Days after treatment	Plant species	mg. Gamma per sq. ft.
1	*Ficus dawei*	4·5
	Culcasia scandens	1·8
	Triumfetta macrophylla ...	2·99
	Alchornea cordata	2·3
	Haemanthus multiflorus ...	1·8
	Aframomum sp.	0·39
11	*Alchornea cordata*	1·17
17	*Alchornea cordata*	1·17
	Triumfetta macrophylla ...	5·07

Laboratory bred flies were exposed to the treated leaves of *Alchornea cordata* (Euphorbiaceae). Kills were negligible on the eighth day and onwards. Results are given in Table II.

TABLE II.

Toxicity to *G. palpalis* of leaves of *Alchornea cordata* from Nfo Island.

Days after treatment	No. of flies	Exposure	Per cent. kill in 24 hours	Per cent. kill in 48 hours
1	15	10 seconds	47	60
	10	Control	10	20
3	15	10 seconds	87	93
	10	Control	10	40
8	15	10 seconds	13	13
	10	Control	0	10
11	17	10 seconds	0	6
	10	Control	0	0
17	15	10 seconds	7	7
	10	Control	0	0

Results.

Selected patches of vegetation amounting to approximately 2 acres of a total of 100 acres were sprayed with a solution of Gammexane in 50 per cent. kerosene and 50 per cent. cottonseed oil. There was a reduction of about 50 per cent. in *G. palpalis* over some 10 days after the treatment. Flies then began to increase in number (fig. 1).

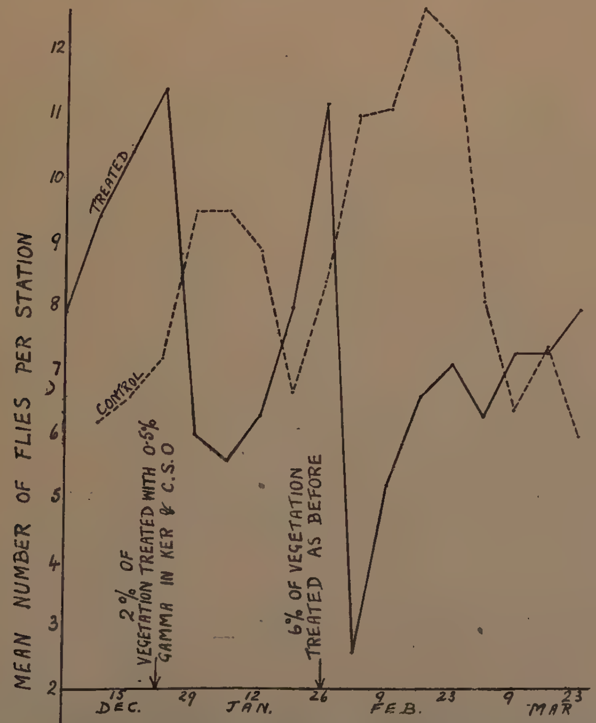


Fig. 1.—Effect of single applications of Gammexane on weekly averages of daily catches of *G. palpalis.*

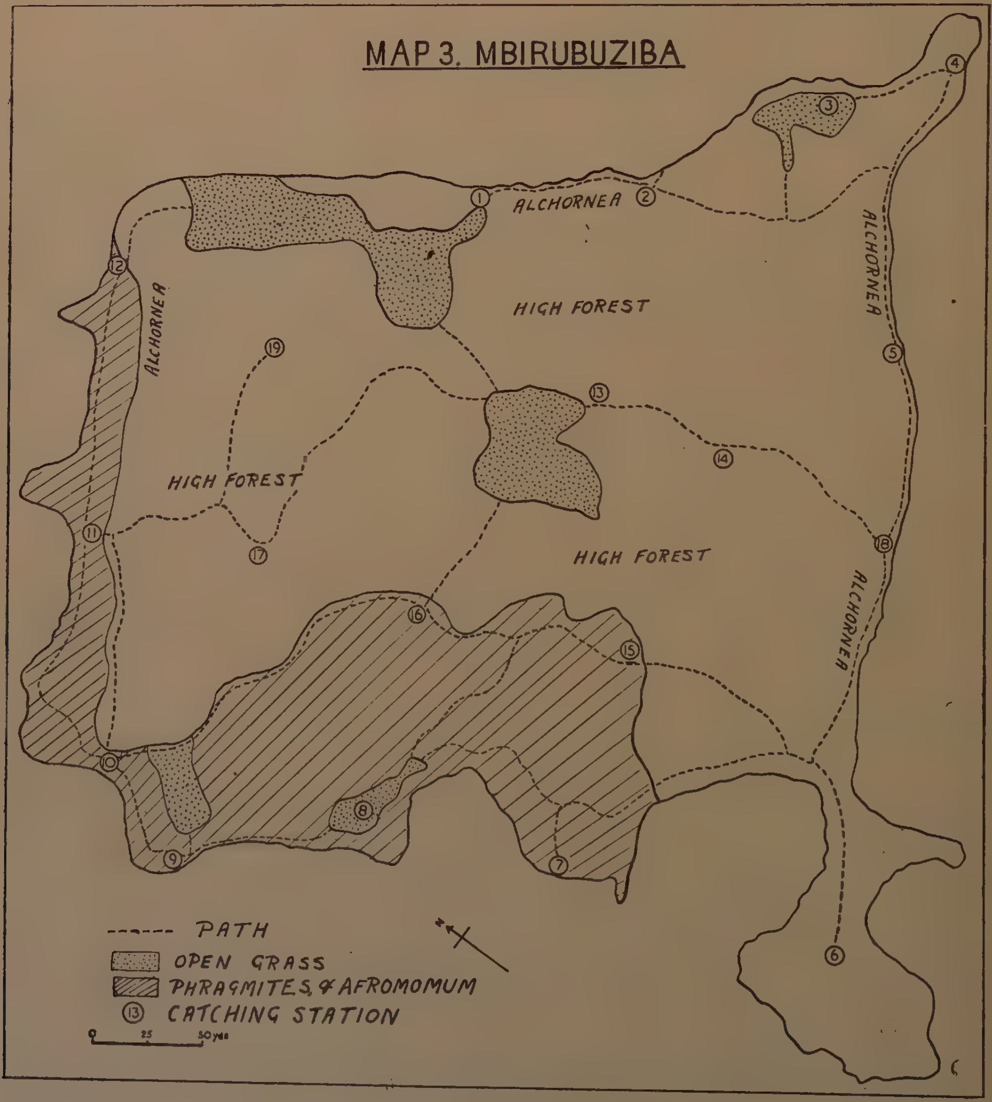
Deposits of Gammexane estimated on the leaf surfaces of samples of treated vegetation were low even one day after the application.

Field Experiment 2. Mbirubuziba Island. (Map 3.)

Mbirubuziba is an island on Lake Victoria, approximately 1 mile SSE. of Mbiru Peninsula and with an approximate area of 30 acres (Map 1).

The island is uniformly flat, nowhere rising to more than 10 feet above lake level. It is nearly surrounded by a belt of reeds in muddy shallow water although the coast proper is rocky.

On the south and east coast there is a narrow belt of ambatch (*Herminiera elaphroxylon*), and *Sesbania sesban*. Behind this there are small sandy clearings


MAP 3. MBIRUBUZIBA
ALCHORNEA
ALCHORNEA
ALCHORNEA
ALCHORNEA
HIGH FOREST
HIGH FOREST
HIGH FOREST
①
②
③
④
⑤
⑥
⑦
⑧
⑨
⑩
⑪
⑫
⑬
⑭
⑮
⑯
⑰
⑱
⑲
PATH
OPEN GRASS
PHRAGMITES & AFROMOMUM
⑬ CATCHING STATION
0
25
50 yds

Map 3.

surrounded by shrubs up to 15 feet high—*Triumfetta macrophylla*, *Alchornea cordata*, *Erlangea tomentosa* (Compositae), and *Aframomum* sp. On the south coast, behind the ambatch, is a wide dense belt of *Phragmites* and *Aframomum*, 10–15 feet high. There is a wide belt of ambatch swamp, on the west coast.

The centre of the island is covered by sparse, high forest with the trees so spaced that there are numerous small clearings to which the sunlight filters producing a mosaic of light and shade on the undergrowth. Trees consist largely of species of *Vangueria* (Rubiaceae), *Canthium vulgare* (Rubiaceae), and *Ficus* spp. The undergrowth is not very dense. It includes *Justicia* sp., *Culcasia scandens*, *Erlangea tomentosa* and *Dracaena* sp.

Mbirubuziba has apparently been frequented by hippo over a long period, for some of their tracks are old and are a distinctive feature of the island. They are seen most frequently on the south and west coasts. Monitor lizards (*Varanus*) occur throughout the island, and crocodiles are seen on the small rocky islands off the south coast. Large colonies of cormorant make use of these rocky islands. Fish eagles, white herons, and ducks are present but not in large numbers.

Dragonflies and lake flies (Chironomids) are abundant, but generally no species of insect occurs in strikingly large numbers.

Fishermen use the island when drying fish.

A few Cassava plants seem to indicate that the island was at one time populated. There is no cultivation now.

Nineteen catching stations were established. Density catches consisting of two fly boys catching for 15 minutes at each station were made daily both before and after treatment.

No control area was set up other than that at Misoli (fig. 1).

Application of Insecticide 29.xii.45.

Insecticide.—DDT manufactured by Geigy, with 83 per cent. para-para content. A concentrate (25 per cent.) was made by dissolving the insecticide in power kerosene warmed to approximately 50°C. over a water bath. This was added to a mixture of kerosene and cottonseed oil to give a 5 per cent. solution of DDT (4·15 per cent. p,p′) in 50 per cent. power kerosene and 50 per cent. cottonseed oil.

Spraying Apparatus.—As for Field Experiment 1.

Dosage.—Total material used—37 gals. of solution. Total spraying time—73½ minutes (from 9.30 a.m. to 3 p.m.). Approximate area covered—74,250 sq. ft. at the rate of 2 qts. per 1,000 sq. ft. or 100 mg. DDT (83 per cent. p,p′) per sq. ft. For the whole island dosage was at the rate of 9·9 oz. DDT (83 per cent. p,p′) per acre.

Area Sprayed.—Areas of vegetation were treated at selected concentration points on the coast, and in the clearings in the centre of the island, especially along hippo tracks; rocks, fallen tree trunks, and dead branches were treated where they occurred in the selected areas.

There was a hot sun, clear sky, and light breeze on the day of application. No meteorological instruments were available on the island.

Initially the insecticide was recovered by simple stripping with benzene but it was later found that drying and powdering, followed by extraction in a soxhlet apparatus, gave a further recovery. This further quantity is evidently from the inside of the leaf. In preliminary work leaves of approximately 30 sq. in. were washed with two lots of 20 cc. of benzene followed by a further 20 ccs. and these two extracts analysed separately. The washings were performed by shaking the whole leaf with the solvent in a stoppered flask for approximately half a minute. It was found that the first extract with two portions of benzene was adequate for all the insecticide preparations used, and also that the relative values found inside and outside the leaf did not depend upon the extent of washing within these limits.

Treated vegetation was collected on the 1st, 10th and 16th days after the application was made. *G. palpalis*, 1 week old and fed on sheep daily, were exposed to these leaves for 10 seconds. Chemical determinations of insecticide on the leaves were then made ; results of these tests are shown in Table III and IV.

TABLE III.

Toxicity to *G. palpalis* of leaves collected from Mbirubuziba Island.

Time after treatment	Leaves used	No. of flies exposed	Exposure	Per cent. kill in 24 hours	Per cent. kill in 48 hours
1 day	*Alchornea*	15	10 secs	66·7	80
	Control	10		0	0
10 days	*Alchornea*	15	10 secs	46·7	60
	Triumfetta	15	10 „	13·3	26·7
	Control	10		10	30
16 days	*Alchornea*	20	10 secs	5	10
	Triumfetta	20	10 „	15	20
	Control	20		0	10

TABLE IV.

Chemical determination of deposits from leaves from Mbirubuziba Island.

Leaves used	Days after spraying	Mg. DDT per square foot	
		From outside by washing	From inside by extraction
Alchornea	1	114 and 111	Not determined
	6	130	31
	6	96	22
	10	58	8
	16	113	29
Triumfetta	10	77	8
	16	34	4

Results.

Vegetation was sprayed with 5 per cent. DDT (83 per cent. p,p′) solution in 50 per cent. power kerosene and 50 per cent. cottonseed oil. 1·7 acres of a total of 30 acres were treated. An immediate reduction of flies by about 80 per cent. seemed apparent. This lasted for about a week then the fly density increased suddenly, and fluctuated somewhat until it reached pre-treatment proportions (fig. 2).

There appears to be an appreciable loss of insecticide through penetration into leaves. Exposures of 10 seconds in laboratory tests are probably too critical.

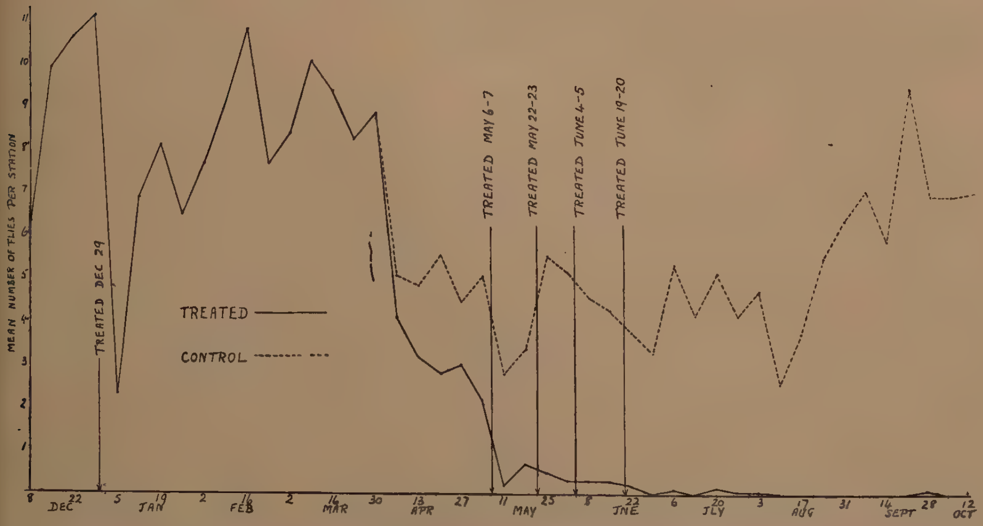


Fig. 2.—Field experiments 2 and 4. Effect of application to vegetation of 5 per cent. DDT in 50 per cent. kerosene and 50 per cent. cottonseed oil on weekly averages of daily captures of *G. palpalis.*

Field Experiment 3. Nfo Island.

By the end of the 4th week after the first spraying on this island (23.xii.45) flies had increased to their pre-treatment numbers. It seemed that the single treatment had failed, particularly against females, probably because the resting or the breeding sites had escaped treatment. For the second experiment it was decided to make a search for pupae, and to make density catches between stations in the hopes of obtaining data on female concentrations.

Considerable numbers of pupae were found in sand between stations 1 and 13. A few were found near 18. Relatively high female rates were found between 4 and 1, 1 and 13, 16 and 18. In the centre of the island the highest female rate was recorded near station 7, where there were fallen tree trunks.

At the initial spraying the lake side of the long coastal belts between 13 and 14, and between 1 and 2 received no treatment. The idea of spraying shore vegetation from a boat was entertained but a survey showed this to be impracticable because of the density of the reeds in places. Additional paths on the lake shore were therefore cut.

A second experiment was made on the 25th and 26th January, 1946. Results of density catches made before and after this second spraying are shown in fig. 1.

Application of Insecticide.

Insecticide.—As in Field Experiment No. 1.

Spraying Apparatus.—As in Field Experiment No. 1.

Dosage.—Total material used—137 gals. Approximate area covered—28,700 sq. yds. at the rate of 2·1 qts. per 1,000 sq. ft. For the whole island dosage was at the rate of 1·4 oz. gamma isomer per acre.

Area sprayed.—Small patches of vegetation round the island were sprayed as before, with additional areas between 13 and 14, 1 and 2, and 16 and 19. Vegetation in the interior of the island was also sprayed. Attempts were made to spray underneath the leaves as well as on the upper surface and the spray was directed into shaded, dense patches of stems and branches.

Density catches, shown in figure 1, were made in the control area on the mainland at Misoli.

During operations there was a hot sun and strong breeze. No instruments were available on the island ; records taken at the mainland, 2 miles away, showed :—

Mean daily maximum temperature °F. for Jan., 83 ; for Feb., 80·9
,, ,, minimum temperature °F. for Jan., 64·5 ; for Feb., 65·1
,, ,, R.H. at 8·30 a.m. for Jan., 77 per cent. ; for Feb., 56 per cent.
,, ,, R.H. at 2.30 p.m. for Jan., 83 per cent. ; for Feb., 70 per cent.

A dry period followed the application until 26.ii.46, when 0·05 in. of rain fell.

No biological or chemical tests with treated vegetation were conducted.

Results.

On Nfo Island the second spraying with a 5 per cent. solution of Gammexane (0·65 per cent. gamma) in 50 per cent. kerosene and 50 per cent. cottonseed oil covered approximately 6 acres of the total area of 100 acres. Reduction of flies by about 80 per cent. occurred over the first week after treatment. Numbers of flies then began to rise slowly and irregularly (fig. 1).

Field Experiment 4. Mbirubuziba Island.

Field experiments 1 to 3 each consisted of a single application of an insecticidal solution to vegetation. In each case there was an immediate marked reduction in fly density as measured by standard captures, lasting for a week to 10 days, followed by a relatively rapid increase and subsequent return to pre-treatment density.

Semi-field and laboratory tests showed a loss of toxicity of deposits on vegetation to flies exposed to them for short periods after 1–2 weeks.

In field experiment No. 4, four applications of insecticide were made at roughly 10 to 14 days intervals in order to maintain an effective deposit over a period equal to the maximum pupal period.

TABLE V.

Patrol Catches—Mbirubuziba Island.

Week	Total number of flies	Number of females	Patrol distance in yards	Mean number of flies per 1,000 yards
Before				
April 22–27	52	12	2,950	17·9
29–4	222	36	8,350	26·6
After				
May 6–11	7	1	4,250	1·6
13–18	12	0	5,300	2·3
20–25	30	8	8,700	3·5
27–1	8	6	5,200	1·5
June 3–8	7	5	5,900	1·2
10–15	4	3	2,950	1·3
17–22	3	0	5,900	0·5
24–29	0	0	8,850	0·0
July 1–6	4	2	8,850	0·4
8–13	1	0	4,600	0·2
15–20	1	0	2,950	0·3

In experiment No. 5, time tables could not be kept so that intervals between applications were not as they should have been and the period of maintained deposit was rather too short.

A description of the island is given in the account of Field Experiment 2.

Density catches were made at established stations, as before, on the first three days of each week. Two boys walking slowly along paths made patrol catches on the remaining three days of each week. This procedure was adopted before treatment and for a month after the fourth application of insecticide. Daily catches were then suspended and the island was visited once a week when density catches were made. Fly captures are given in fig. 2 and Table V.

It is regretted, for various reasons, that standard catches could not be made at all stations or on all patrols each day of the week.

Density and patrol catches, shown in fig. 2 and Table VI, were made on the mainland at Mbiru, approximately 1 mile from the island (Map 1).

By the usual method of ageing flies, based on the softness of the cuticle, samples of flies were divided into two age-groups—"young" or up to 7 days old, and "old" over 7 days. Numbers recorded before and after treatment are shown in Table VII. Since the staff were not very experienced in this work the figures were probably not very accurate.

TABLE VI.

Patrol Catches—Mbiru Mainland.

Week	Total number of flies	Number of females	Patrol distance in yards	Mean number of flies per 1,000 yards
April 22–27 ...	144	37	2,340	61·5
29–4 ...	82	14	2,520	32·5
May 6–11 ...	32	4	2,120	15·1
13–18 ...	95	9	4,680	20·3
20–25 ...	139	11	4,180	33·2
27–1 ...	70	9	3,110	22·5
June 3–8 ...	124	12	5,040	24·6
10–15 ...	108	7	5,040	21·4
17–22 ...	154	34	7,560	20·4
24–29 ...	178	45	7,560	23·5
July 1–6 ...	126	30	7,560	16·7
8–13 ...	194	74	7,560	25·7
15–20 ...	143	51	7,560	18·9

TABLE VII.

Age of samples of flies on Mbirubuziba Island.

	Total number of flies examined	Young (up to 7 days)		Old (over 7 days)		Per cent. young flies
		Male	Female	Male	Female	
9.4.46–18.4.46, before spraying ...	334	110	24	119	81	40
16.5.46–23.5.46, after 1st spraying	76	41	14	13	8	72
24.5.46–4.6.46, after 2nd spraying	51	14	20	5	12	67
6.6.46–22.6.46, after 3rd spraying	26	10	13	1	2	88
23.6.46–13.7.46	9	5	3	0	1	89

Application of Insecticide.

Insecticide.—DDT solution in 50 per cent. power kerosene and 50 per cent. cottonseed oil. DDT of 83 per cent. p,p′ isomer was used throughout. For the first spraying the solution was made up to contain 4·15 per cent. pure DDT; it contained 5 per cent. para-para DDT for the other applications.

Spraying Apparatus.—Four Oaks "Kent" Sprayer fitted with 2 nozzles for the first application. It was considered that this was too wetting, and for subsequent applications only one nozzle was used. At an average rate of progress this covered 230 sq. yards in 3 minutes.

Dosage.—The dosages applied were as given below.

	Insecticide used	Dosage per acre for whole island
1st spraying 6.5 and 7.5.46 ...	96 gallons	1·33 lbs. pure DDT
2nd ,, 22.5 and 23.5.46	38 ,,	0·63 ,, ,,
3rd ,, 4.6 and 5.6.46 ...	40 ,,	0·67 ,, ,,
4th ,, 19.6. and 21.6.46	81 ,,	1·35 ,, ,,

Areas Treated.—Insecticide was applied at selected areas along the coast and in the centre of the island, along hippo tracks where fly movements had been noted, on prominent headlands, at suspected breeding places where unhatched pupae had been found, at probable resting places and under fallen tree trunks—in all about 4 acres.

Labour.—Labour for the first spraying consisted of two squads; for the others only one squad. Each squad consisted of a senior boy to supervise, two boys to fill sprayers and maintain pressure, one boy to spray, and one porter to carry tins of solution.

Cost.—

	Shs. cents
Labour	83/00
144 lb. DDT (83 per cent. p,p′) at 4 Shs. 70 c. per lb.	676/80
127·5 gals. power kerosene at 1/54 per gal.	196/35
127·5 gals. cottonseed oil at 3/82 per gal.	487/05
Total	1443/20
Cost per acre over whole island	48/10

A meteorological station was established on the island. Records are given in Table VIII. From the first day of spraying (6.v.46) until 20.vii.46, four weeks after the last spraying, 317·7 mms. of rain fell in 52 showers of which 38 were heavy and of more than one hour duration. The deposits therefore had to withstand fairly heavy washing by rain.

The method of sampling treated vegetation in previous trials was perhaps too crude. To improve on this a small patch of *Alchornea* bush and another of *Triumfetta* on the island were carefully sprayed once only to give as even a coverage as possible. Branches were brought into the laboratory at various times after the application. Two leaves were taken from each branch at random and 10 flies exposed to each for 20 seconds. Chemical determinations of the deposits on the surface of the leaves, and the amount inside the leaves were then made.

TABLE VIII.

Meteorology—Mbirubuziba Island.

Week ending	Temperature °F.		Percentage Humidity			Rainfall	
	Max.	Min.	7.30 a.m.	12 noon	6 p.m.	Amount in mms.	Number of showers
20.4.46	76·8	65·0	91·3	79·1	76·6	69·5	6
27.4.46	78·2	65·6	88·5	73·5	78·7	30·5	2
4.5.46	77·3	66·0	92·3	77·7	76·5	32·2	5
11.5.46	77·0	67·1	91·5	78·5	83·5	13·7	5
18.5.46	79·0	67·7	91·4	75·8	75·9	46·0	3
25.5.46	76·7	64·9	88·2	79·7	83·8	25·4	8
1.6.46	70·3	66·3	95·5	83·8	84·8	75·2	8
8.6.46	75·6	66·6	86·4	80·4	81·8	6·8	3
15.6.46	72·8	64·5	94·1	87·2	82·8	119·0	16
22.6.46	75·8	64·6	94·3	77·3	80·3	23·0	4
29.6.46	74·2	64·6	88·3	77·6	82·0	3·0	2
6.7.46	76·5	64·2	87·0	74·0	82·0	0·0	0
13.7.46	78·8	65·0	91·0	66·0	81·0	2·0	2
20.7.46	81·0	64·3	85·0	72·0	—	3·6	1
27.7.46	77·2	63·3	96·0	75·0	86·0	12·0	2

Flies used were laboratory bred *G. palpalis*, fed daily on sheep, and 7 to 10 days old at the time of the test. Figures for controls are not given, as deaths were negligible.

For the chemical estimations leaves were first stripped with benzene to remove surface deposits. They were then dried, powdered and extracted in a soxhlet apparatus for the absorbed insecticide.

A solvent such as benzene will penetrate into the leaf during washing and remove interior DDT. To eliminate errors from this source the washing procedure was standardised by shaking the whole leaves with a given volume of solvent for a given time. A deposit entirely on the outside of the leaf can be obtained by slowly applying a known volume of DDT solution in ether from a pipette and directing a stream of air over the leaf so that the ether evaporates almost as soon as it touches the surface. Preliminary experiments showed that such a deposit on a leaf of area 30 sq. inches was completely removed by shaking with 20 ml. benzene for half a minute followed by a further 20 ml. for half a minute.

This procedure was adopted for all leaves of area up to 30 sq. inches (two washings of 30 ml. each were used for larger areas). DDT not removed by this washing but found in a soxhlet extract was considered to have been inside the leaf.

Results.

(i) Fly activity upon which density catches are based, varies markedly with changes of weather. Weekly means, therefore, fluctuate. It was originally intended to commence the first spraying for this experiment during the first week in April. The rains started at this time and operations were postponed. The many dull or wet days that followed had a marked effect both at Mbirubuziba and Mbiru; fly densities were more than halved (fig. 2). The wet weather continued and although fly densities were very low, it was decided to start operations.

(ii) As a result of the first spraying mean weekly density catches fell from 2·2 flies per station on 4th May to 0·2 per station on 11th May. One week later the figure had risen to 0·7. Following each subsequent spraying there was a noticeable reduction, and a slight increase from a few days after the spraying until the next application.

After the fourth spraying no flies were seen for several days; then one or two were caught each day. From the 6th to the 13th week after the final application no flies were seen or caught on the fly rounds; then two flies were caught on 26.ix.46 (fig. 2).

These results were supported by patrol catches for the corresponding period.

(iii) On Mbiru Mainland, which was used as a control, the density and patrol catches remained fairly constant over the same period (fig. 2 and Table VI).

(iv) A factor that may have interfered somewhat with fly reduction on the island is the movement of fishermen. It has been impossible to keep them out of this experimental area, and from experience it is known that canoes frequently convey tsetses on them for considerable distances. It may well be that flies occasionally reached the island by this means after the final spraying.

(v) In the sample of flies aged before spraying commenced 60 per cent. were more than a week old. The numbers of flies caught after the 2nd, 3rd and 4th spraying were too low to permit estimation of the proportion of "old" flies. However, it appears that some "old" flies escaped the action of the insecticide (Table VII).

(vi) Chemical laboratory tests conducted with samples of treated vegetation showed that there was considerable initial loss of insecticide by penetration into the leaves—as much as 50 per cent. in the case of the soft leaved *Triumfetta*. Loss of insecticide from the surface of leaves continued, until 5 weeks after the application only 13 mg. per sq. ft. remained. During this period the deposits had to withstand heavy washing by rainfall (Table IX).

Biological tests showed a general decrease (from 100 per cent. to 5 per cent. over the 5 weeks) in toxicity to *G. palpalis* exposed for 20 seconds contact (Table IX).

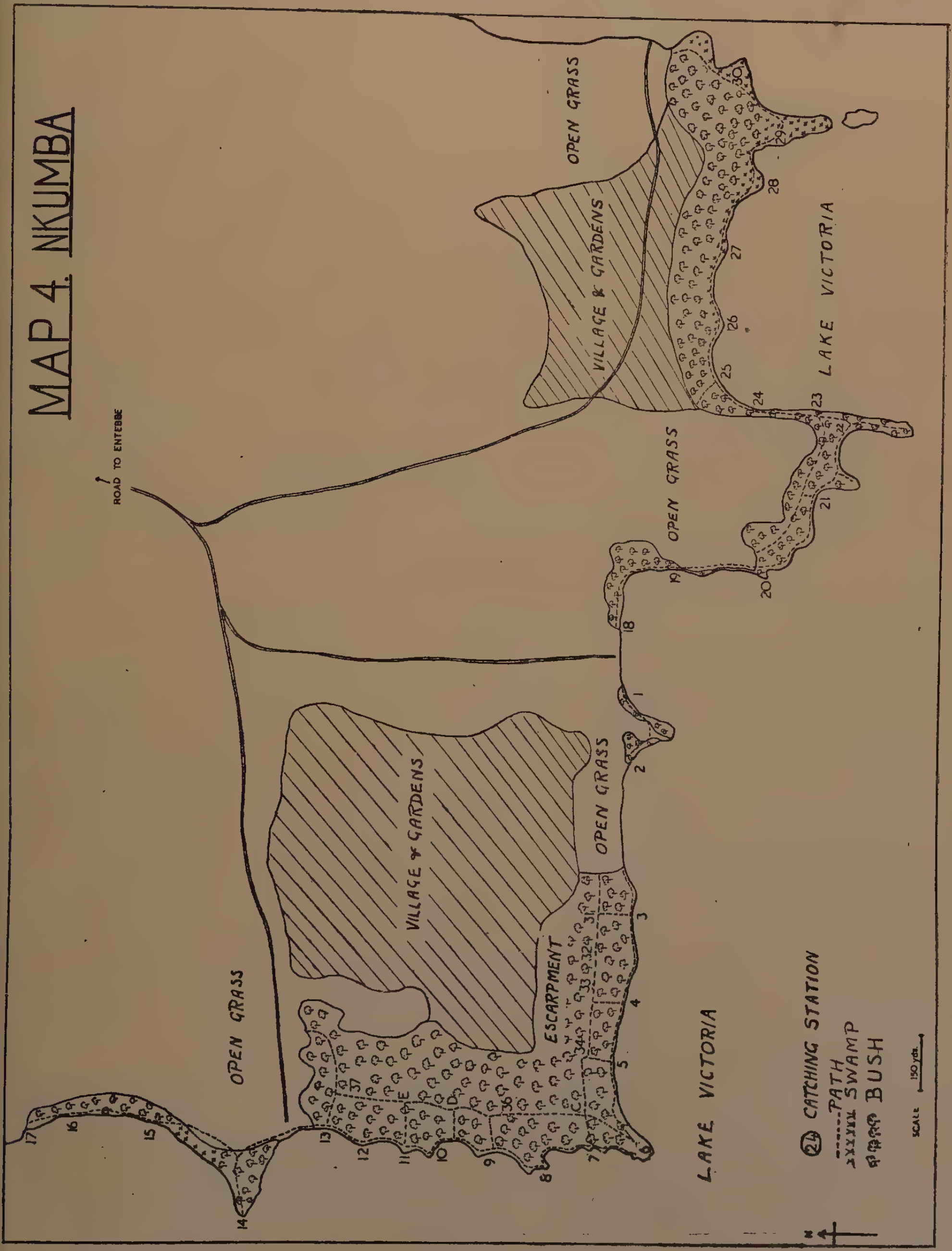
TABLE IX.

Toxicity to *G. palpalis* of leaves treated with 5 per cent. DDT solution in 50 per cent. power kerosene and 50 per cent. cottonseed oil on Mbirubuziba Island, and recovery of DDT from the same leaves.
(Control deaths were negligible: figures are not given.)

Leaves used	Number of days after spraying	Recovery of DDT in mg./sq. ft.		Per cent. kill of *G. palpalis* 24 hours after exposure of 20 seconds
		Outside of leaves by washing	Inside of leaves by extraction	
Alchornea	1	100	59	100
	3	66	50	68
	5	72	29	35
	8	80	35	25
	13	48	28	25
	21	28	12	10
	25	22	8	25
	28	14	7	25
	35	13	9	10
Triumfetta	1	61	65	45
	5	60	26	40
	8	60	11	15
	13	60	7	10
	21	92	15	20
	25	23	2	30
	28	47	7	15
	35	29	4	5

Field Experiment 5.—Nkumba. (Map 4).

Nkumba is a small peninsula of the mainland a few miles east of Entebbe (Map 1). The experimental area is confined to lake shore bush on the west and south coasts; and for convenience is divided into two parts, " west " (catching stations 1–17) and " east " (catching stations 18–30). The lake shore, with its bush, has a coast line of 3 miles, and an area of approximately 130 acres (Map 4).



MAP 4. NKUMBA
ROAD TO ENTEBBE
OPEN GRASS
VILLAGE & GARDENS
OPEN GRASS
LAKE VICTORIA
OPEN GRASS
VILLAGE & GARDENS
OPEN GRASS
ESCARPMENT
LAKE VICTORIA
OPEN GRASS
24 CATCHING STATION
PATH
SWAMP
BUSH
SCALE 150 yds.
N

Map 4.

The "west" area is limited in the north by a man-made clearing separating it from the Entebbe Township. From catching station 17 to 14 there is a narrow strip (750 yards by 50 yards wide) of swamp surrounded by shrubs (*Alchornea*). This is separated by open grassland from the main bush which extends along the rest of the west coast, and for 750 yds. of the south coast. On the west, the edge of the lake is generally swampy, but rocky in some places, with a wide belt of *Alchornea*, up to 15 ft. high, often forming a mass of tangled stems and branches. Behind this, to a depth of approximately 300 yds., is sparse high forest (*Vangueria* spp., *Ficus* spp. etc.) with an undergrowth of *Haemanthus multiflorus*, *Culcasia scandens*, *Justicia* sp., *Dracaena* sp. Between the lake and the belt of *Alchornea* on the south shore is a sandy beach up to 15 ft. in width. The forest here is only 100–150 yds. in depth, being limited by a steep escarpment.

The two main areas are separated by two sandy beaches and a central headland which rises 50 ft. above lake level and bears vegetation (stations 1 and 2).

The "east" area, from catching station 18 to 19, has a width of 30 yds. and consists mostly of *Alchornea* with a swampy lake shore. From catching station 19 to 20 there is a very narrow belt of low shrub; 20 to 23 has a lake shore belt of *Alchornea* with sparse high forest behind; 23 to 25 is a very narrow belt of bush with a very rocky coast line and is limited by a steep escarpment. The remainder of the area, from 25 to 30, has a wide swamp between the forest and the lake shore. The swamp consists mostly of Ambatch and *Phragmites*, and small grassy open spaces surrounded by low shrubs (*Melanthera*, *Triumfetta*). It is traversed by numerous hippo tracks.

The vast majority of leaves have a smooth surface. The dominant species of the lake shore is *Alchornea cordata*. Of the plants with soft, hairy leaves, *Triumfetta macrophylla* and *Melanthera brownei* are the most common.

Monkeys (*Cercopithecus*) are numerous in the forest of the west area. Tracks of hippos are specially numerous on the west shore from catching station 6 to 11, and

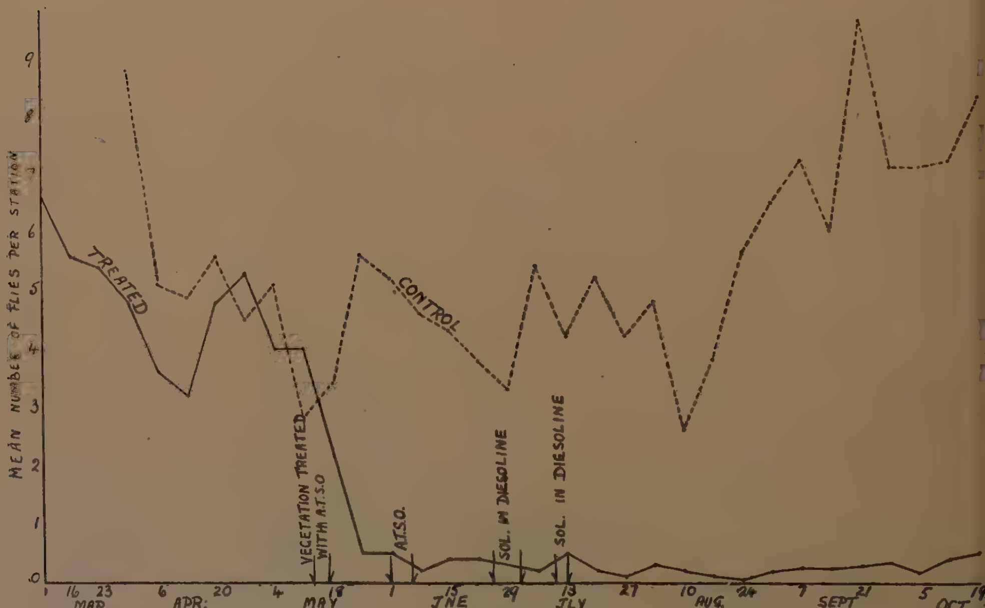


Fig. 3.—Field Experiment 5. Influence of four applications of 5 per cent. DDT on weekly averages of daily catches of *G. palpalis*.

on the south shore from 23 to 29. Few crocodiles have been seen, but monitor lizards (*Varanus*) are common.

Immediately behind each main area is a village of native huts among banana plantations. There is continuous traffic of water carriers between the villages and the lake. Fish is brought daily to the sandy beach between catching stations 1 and 18, where large numbers of Africans congregate.

G. palpalis is present in the lake shore bush from catching station 1 to 30. Movement of fly between the east and the west areas has been shown by fly-marking and subsequent recaptures.

Catching stations were established after paths had been cut. Density catches were made on the first three days of each week ; patrol catches on the remaining three days of each week. Records before and after treatment are given in fig. 3 and Table X.

TABLE X.

Patrol Catches—Nkumba.

Week	Total number of flies	Number of females	Patrol distance in yards	Mean number of flies per 1,000 yards
Before				
Mar. 4–9 ...	446	166	14,150	31·5
11–16 ...	531	207	16,650	31·9
18–23 ...	640	252	18,750	34·1
25–30 ...	—	—	—	—
Apr. 1–6 ...	334	109	16,200	20·6
8–13 ...	391	155	14,600	26·8
15–20 ...	103	33	4,900	21·0
22–27 ...	295	72	13,000	12·7
29–4 ...	445	165	18,900	23·5
May 6–11 ...	338	54	16,350	20·7
After				
May 13–18 ...	81	25	16,950	4·8
20–25 ...	50	13	16,050	3·1
27–1 ...	33	4	14,400	2·3
June 3–8 ...	34	8	9,650	3·5
10–15 ...	6	1	5,600	1·1
17–22 ...	32	13	14,550	2·2
24–29 ...	—	—	—	—
July 1–6 ...	40	15	14,200	2·8
8–13 ...	17	5	12,650	1·3
15–20 ...	10	4	14,200	0·7
22–27 ...	3	2	9,900	0·3
29–3 ...	5	1	4,950	1·0
Aug. 5–10 ...	8	2	4,600	1·7
12–17 ...	13	5	4,600	2·8
19–24 ...	6	1	4,950	1·2
26–31 ...	12	3	4,950	2·4
Sept. 2–7 ...	9	1	4,950	1·8
9–14 ...	12	3	4,900	2·4

Control.

Mbiru Mainland was used as a control area for this experiment as well as for the previous one (see fig. 3 and Table VI).

Samples of flies were aged as in the previous experiment. Proportions of " young " and " old " flies are given in Table XI.

TABLE XI.

Age of samples of flies at Nkumba.

	Number of flies in sample	" Young " flies (up to about 7 days)		" Old " flies (over about 7 days)	
		Male	Female	Male	Female
Before spraying	195	40	8	130	17
After 1st application	7	2	3	1	1
After 2nd application	13	1	3	7	2

Application of Insecticide.

Four applications were made.

Insecticide.—The first and second applications were made with a 5 per cent. DDT (pure) emulsion, made by adding one part of ATSO concentrate to two parts of lake water. Supplies of ATSO were then exhausted and for the third and fourth sprayings a 5 per cent. (pure) DDT solution in dieseline was used.

Concentrated ATSO I supplied by the Ministry of Supply consists of :—

Heavy solvent naphtha	3 gals.
DDT	7 lb.

with emulsifier of

Turkey-red oil	50 per cent.	1 gal.
m-cresol	30 per cent.	
Sodium oleate	15 per cent.	
Water	5 per cent.	

Spraying Apparatus.—Four Oaks " Kent " Sprayers each fitted with one nozzle for the first three sprayings ; an Aerostyle motor sprayer for the fourth. The Aerostyle motor sprayer consisted of a three-wheeled chassis, on which were mounted a J.A.P., $1\frac{1}{2}$ h.p. engine and pump, combined, and a four-gallon container. The pump and engine, connected by a direct drive, could be lifted off the chassis, for carrying where the ground was too rough for wheeling the chassis. A twelve-yard length of hose was fitted, so that approximately 25 yards of vegetation could be sprayed without moving the outfit. Solution was brought to the sprayer in four-gallon tins from which it was possible to spray directly, with a considerable saving in time.

Dosage.—The dosages applied were as given below.

	Volume of insecticide	Dosage per acre for whole area.
1st spraying 13.5.46 ATSO emulsion	150 gallons	0·58 lbs. pure DDT
2nd ,, 29.5.–6.6.46 ATSO emulsion	108 ,,	0·41 ,, ,, ,,
3rd ,, 25.6.–2.7.46 DDT solution in dieseline	156 ,,	0·6 ,, ,, ,,
4th ,, 8.–11.7.46 DDT solution in dieseline ...	96 ,,	0·37 ,, ,, ,,

Fifteen sample papers fixed to leaves during the third spraying gave an average deposit of 44 mg. DDT per sq. ft. Papers at 5–6 ft. had 25 mg., those at 2–3 ft. had 87 mg. and at 1 ft. 20 mg.

Areas treated.—Vegetation was treated to a height of about 6 ft. along the lake shore (Pl. XI, fig. 1). At some places, where the path was not at the lake edge, vegetation on both sides of the path was sprayed at the first spraying. Vegetation on the lake side of the inner paths from 37 via C to 31, and from 20 to 23 was also treated. A linear distance of approximately 9,000 yds. was sprayed. Applications were confined to the lake shore in the 2nd, 3rd and 4th sprayings, a linear distance of about 6,000 yds.

Effect on Vegetation.—Some of the emulsion drained off the smooth surfaced leaves. After evaporation of the water, a matter of a few hours, the leaf surfaces felt " tacky ". Volatilisation of the oil phase followed, leaving a white, partially crystalline deposit on the leaf surfaces. No burning of the glabrous leaves was obvious. Very little deposit could be seen on the soft hairy leaves. The dieseline solution causes burning of some soft leaves and the tips of some of the harder species.

Labour.—For the first spraying, labour was divided into two squads each consisting of a supervisor, one boy to spray, two boys to make up the emulsion from the concentrate and lake water, and to serve the sprayers, and one porter. Spraying was spread over three days, both squads working for 6 hours each day.

For the second and third applications one squad only was used. The motor sprayer was used for the fourth application. Two boys carried the engine and maintained it, one boy worked the spray gun, one helped with the hose and a porter did general carrying.

TABLE XII.

Meteorological Records at Entebbe.

Week ending	Temperature °F.		Percentage Humidity			Rainfall	
	Max.	Min.	6 a.m.	12 noon	6 p.m.	Total inches	Number of days
30.3.46	—	—	—	—	—	0·15	2
6.4.46	—	—	—	—	—	4·64	5
13.4.46	79	64	—	—	—	4·13	6
20.4.46	78·0	64·0	—	—	—	2·89	6
27.4.46	78·0	62·0	—	—	—	1·30	3
4.5.46	77·5	62·0	93	84	85	5·17	5
11.5.46	78·0	61·0	97	85	64	2·54	4
18.5.46	79·5	64·0	94	83	83	1·77	4
25.5.46	77·0	64·0	93	81	80	2·25	5
1.6.46	78·0	62·0	93	86	77	2·33	4
8·6·46	78·0	63·0	91	79	80	1·18	2
15.6.46	76·0	62·5	95	87	83	3·96	6
22.6.46	76·5	61·5	92	82	79	1·48	5
29.6.46	76·0	64·0	93	85	78	0·71	2
6.7.46	79·0	63·0	92	73	80	0·0	0
13.7.46	75·5	64·0	94	81	78	0·17	1
20.7.46	77·0	64·0	95	76	79	0·19	2
27.7.46	74·5	63·0	95	79	78	1·77	5
3.8.46	74·5	63·5	97	79	79	0·29	4
10.8.46	75·0	62·5	95	77	81	1·02	4
17.8.46	73·5	62·5	96	81	86	1·46	5
24.8.46	75·0	63·0	96	78	85	0·01	1

Cost of Spraying.—The price of ATSO concentrate is unknown so that the cost is given only for the third and fourth sprayings.

	Shs.	Cts.
Labour	58	00
DDT 151 lb. (83 per cent. p,p′) at 4 Shs. 70 c. a lb.	709	70
Dieseline, 252 gallons at 0/89 per gallon	224	28
Total	991	98
Cost per acre for whole area (for 2 applications)	7	63

Operations were often interrupted by rain. Very heavy rain fell during the night following the first day's spraying, and there was a shower lasting ten minutes just before the completion of the first spraying. Operations were suspended for four days during the second spraying, and for three days during the third spraying because of rain. Details of meteorological records kept at Entebbe are given in Table XII. Between 13.v.46, the first day of spraying, and 26.vii.46, two weeks after the final application, 18·35 inches of rain fell. There were many heavy storms.

A small area of bush was specially treated, once only, with emulsion during the first spraying, and samples of leaves were taken at intervals for laboratory tests. Another small area was given one application of the DDT/Dieseline solution. Sampling and techniques were as described for Field Experiment 4. Results are given in Tables XIII and XIV.

TABLE XIII.

Toxicity to *G. palpalis*, exposed for 20 seconds contact, of leaves treated at Nkumba with 5 per cent. DDT emulsion (ATSO), and recovery of DDT from the same leaves.

Leaves used	Age of deposit	Per cent. kill of *G. palpalis* in 24 hours	Recovery of DDT mg./sq. ft.	
			On surface of leaves	Inside by extraction
Alchornea	3 days	25	33	16
	6 "	40	49	1
	8 "	53	34	1
	11 "	60	38	8
	11 "	40	21	8
	18 "	60	Not determined	Not determined
	8 weeks	65	42	" "
	16 "	10	10	3
	16 "	0	12	10
Triumfetta	3 days	0	50	5
	8 "	0	54	5
	18 "	10	Not determined	Not determined
Dracaena	8 weeks	100	116	Not determined
	16 "	93	173	16

TABLE XIV.

Toxicity to *G. palpalis*, exposed for 20 seconds contact, of *Alchornea* leaves treated at Nkumba with 5 per cent. DDT solution in dieseline, and recovery of DDT from the same leaves.

Age of deposit	Per cent. kill *G. palpalis* in 24 hours	Recovery of DDT mg./sq. ft.	
		On surface of leaves	Inside by extraction
1 day ...	75	(?)	33
4 days ...	80	62	10
8 „ ...	35	12	9
14 „ ...	20	17	10
26 „ ...	10	7	17

Results and General Observations.

(i) As in the previous trial, fly activity and therefore density catches were influenced considerably by the heavy rains which began at the beginning of April; figures before treatment were not as large as had been hoped.

Rain also interfered with spraying operations and it was not possible to make the applications at regular intervals. Periods between sprayings were 14, 18 and 6 days, and the total period of probable maintained deposit—48 days—was too short.

(ii) Over the whole area, fly densities were reduced by about 98 per cent. as a result of the four applications (see fig. 3). Patrol catches during the corresponding period confirm these results (see Table X).

(iii) Fly densities were always higher on the " east " side than on the " west ". For example, for weeks ending March 9, 16, and 23, densities on the " east " were 7·8, 8·8 and 7·9; those on the " west " were 5·9, 4·4 and 4·5.

After the final application of insecticide flies were rarely caught on the " west " side—one or two each week.

A week after the final spraying flies began to appear in fair numbers at catching stations 29 and 30. It was discovered that villagers had started to move cattle from fly infested bush north of catching station 30, across the clearing to the bush at catching stations 29 and 30; by marking flies it was established that they were coming or were being brought across to the experimental area. This somewhat interfered with the apparent results.

(iv) At Mbiru, the control area, fly densities remained fairly constant at 4 to 5 flies per station (in 15 minutes) throughout the period of the experiment.

(v) During the two weeks prior to the first spraying 75 per cent. of a sample of flies caught at Nkumba were more than 7 days old. After the first and second sprayings numbers of flies were too small to give an estimation of percentage " old " flies, but it was seen that a number of old flies escaped the treatment.

(vi) Insecticidal deposits, as determined from sample papers and from leaves for laboratory tests, were rather small. Deposits on leaves from the ATSO emulsion persisted for several weeks despite heavy storms, and gave fairly good kills in the laboratory. Leaves were taken from a heavily treated *Dracaena* plant which was permanently in shade. Surface deposits of 116 and 173 mg. per sq. ft. gave kills of 100 per cent. and 93 per cent., respectively, 8 and 16 weeks after application of insecticide.

Deposits on hairy leaves gave negligible kills. It may be that the hairs prevent contact between the tarsi of the flies and the deposit.

(vii) Growth of vegetation is very rapid during and immediately following the rainy season. This influences the effectiveness of deposits on leaves in that the proportion of treated leaves, and consequently the probability of a fly alighting on a treated leaf, is considerably reduced. Furthermore, the new growth tends to cover and hide the older treated leaves.

General Conclusions.

Single applications of these insecticidal solutions to a very small proportion of total infested vegetation by hand spraying have resulted in a reduction of 50–80 per cent. of *G. palpalis* for 1–2 weeks only. Repeated applications of insecticide to maintain a toxic deposit over a period of 50–60 days have given a greater and more permanent reduction. It seems, however, that this laborious method of application, even with excessive dosages, is not likely to eliminate *palpalis*.

It should be emphasised that a very thorough study of the fly was not made in any of these experiments, and that only a relatively small proportion of the vegetation was treated. It is obvious that insecticide was not applied to all the vital places, such as female resting haunts.

In a very small fly belt it would be possible after prolonged and detailed study to treat, in this manner, the large majority of fly resting places and patrol routes and so most probably to eliminate fly. But to do this in extensive fly belts would be a tremendous task.

It seems that effort must be devoted to the production and trial of formulations that will not be absorbed by vegetation, that will not lose toxicity during prolonged exposure, and at the same time devise some method of application that will give a much greater coverage of vegetation than can be achieved with small knapsack sprayers.

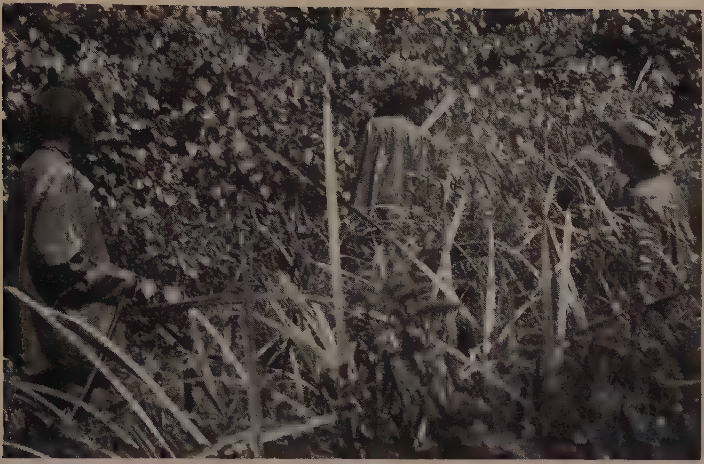


Fig. 1. Spraying lake shore vegetation (Nkumba).

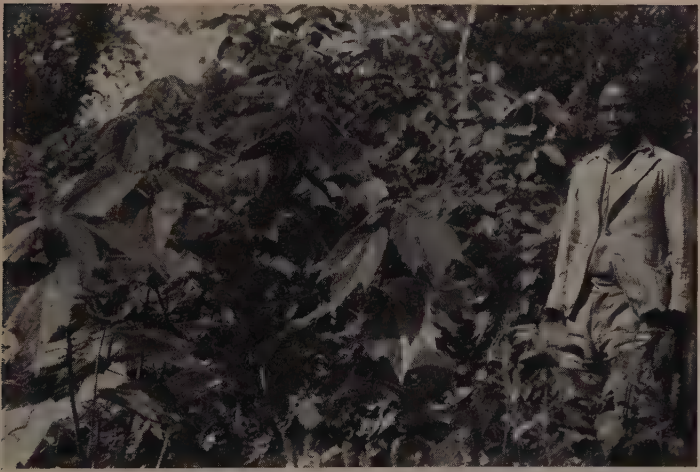


Fig. 2. *Triumfetta macrophylla.*

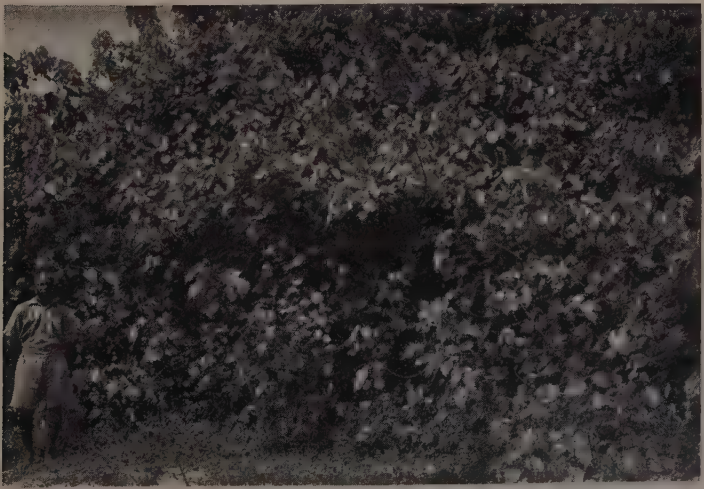


Fig. 3. *Alchornea cordata.*

THE STATUS OF *DACUS ARMATUS* FABRICIUS AND OF *DACUS BIVITTATUS* (BIGOT) (TRYPETIDAE, DIPTERA).

By H. K. Munro, D.Sc., F.R.E.S.
Division of Entomology, Pretoria.

A full consideration of the systematic position of *Dacus armatus* F. (the genotype of *Dacus*) and of *Dacus bivittatus* (Big.) is not attempted here, but it is necessary, owing to the economic importance of the latter, to clear up the confusion that has arisen between them. I am indebted to and thank Dr. A. J. Hesse, South African Museum, Capetown; Dr. John Smart, British Museum; Sir Guy A. K. Marshall, K.C.M.G., and Dr. F. van Emden, Imperial Institute of Entomology, and Dr. S. L. Tuxen, Zoological Museum, Copenhagen, for help in unravelling the problem.

The two species belong to the group of Dacinae (mostly African) in which the terga of the abdomen are fused to form a syntergum as opposed to that (mostly Oriental, some African) in which the terga are free. They are large species, 8–10 mm. *Head:* face with a pair of rather large, black spots; slight, fine, dark pubescence on frons, one upper and two lower orbitals, no ocellars, inner and outer verticals present, but no postorbitals. *Thorax:* four well-developed scapulars, three supra-alars, two scutellars, two notopleurals, one mesopleural and a weak pteropleural; yellow markings strong, on dorsum three postsutural stripes, humeri partly or entirely yellow, mesopleural stripe wide, from notopleural callus to sternum, two hypopleural spots, contiguous or narrowly separated, scutellum yellow, narrowly ferruginous at base. *Wing:* anal stripe strong, the wide costal border includes the upper half of first posterior cell and may extend to fourth vein or barely below it; a distinct, oval, apical patch is not developed. Third abdominal tergum in male ciliate.

The forms may be distinguished as follows:—

1. Hind femora quite yellow; hyaline part of wing distinctly but faintly infuscated, especially rather broadly on the hind margin..........................2

— Hind femora ferruginous on outer third; hyaline part of wing glass-clear as a rule 3

2. The yellow of the pair of hypopleural spots confluent.........*armatus armatus* F.

— The hypopleural spots separated by a narrow, ferruginous stripe, the posterior the smaller..*armatus lulongaensis* Collart

3. Hypopleural spots confluent, or barely a trace of darkening between; normally coloured specimens, ferruginous to chestnut, or with only a tendency to blackness..4

— Distinctly black specimens in which the hypopleural spots are separated by a dark stripe; in male, ejaculatory apodeme wide and phallic apodeme long ....*bivittatus cucumarius* form *atrescens* **nov.**

4. Brown fore-border of wing extends only to middle of first posterior cell; ejaculatory apodeme narrow or but little widened, the phallic apodeme short, about half length of fultella; specimens from West Africa and Gold Coast area...*bivittatus bivittatus* (Big.)

— Brown fore-border extends to fourth vein at least as a not too faint infuscation in the lower part of first posterior cell; ejaculatory apodeme wide and fan-like, phallic apodeme long, about two-thirds length of fultella; specimens from Central, East and South Africa........................*bivittatus cucumarius* Sack

Dacus (Dacus) armatus armatus F.

Fabricius, 1805, Syst. Antl., 273 (*Dacus*).

Wiedemann, 1830, Aussereur. zweiflug Ins., 2 : 517 (*Dacus*).

Becker, 1903, Mitt. zool. Mus. Berlin, **2** p. 131 (*Dacus*).

Bezzi, 1908, Boll. Soc. ent. ital., **39** pp. 138, 147 ; 1908, Boll. Lab. Zool. Portici, **3** pp. 292, 297 (*Dacus*) ; 1915, Bull. ent. Res., **6** pp. 89, 91, fig. 5 ; 1924, *ibid.*, **15** p. 82 (*Tridacus*).

Sack, 1908, Ber. senckenb. naturf. Ges., **1908** p. 8 (*Dacus*).

Froggatt, 1909, Report on parasitic and injurious insects. Dep. Agric. N.S.W. 1907–08 p. 98 (*Dacus*).

Speiser, 1910, Wiss. Z. Exp. Kilim.-Meru, **10** p. 183 (*Dacus*).

Perkins, 1937, Proc. roy. Soc. Qd, **48** p. 52 (*Dacus*).

The following description is based on the female seen by Bezzi and on two males to be noted later.

♂♀. Length 10·3 mm., wing 8·6 mm. The smaller male, length 8·9 mm. *Head:* of normal shape, mainly yellow, brownish behind with a rather wide postocular margin ; comb fine and yellowish ; *frons*, width 0·6 length and 0·32 width of head, yellowish-brown more yellow on sides, the flat median tubercle brown and three, brown, subintegumentary spots on either side ; *lunule* short, brown ; *antennae* blackish-brown, elongate, about 0·3 longer than narrow parafacials, arista bare, base brownish, flagellum black ; *face* yellow, the pair of large black spots drop-shaped in female, the point touching epistome margin, in male more rounded or with a broader, shorter point not reaching epistome ; gena slightly wider than third antennal joint and with a brown spot below eye, bristle short ; palpi normal, proboscis short. *Thorax:* blackish on dorsum with narrow median and dorso-central ferruginous stripes anteriorly and ferruginous on sides, the black is slightly white-dusted and this on the pleura gives rise to the " weisslich schimmernde Binde " mentioned in the original description ; yellow marks strong : humeri, strong postsutural stripes, the lateral ones from yellow spot on suture to just before inner post-alar bristle, the mesopleural stripe normal and hypopleural spot double, scutellum also yellow ; pubescence pale, rather gold-shining, especially before scutellum ; squamae brownish narrow ; halteres yellow ; postscutellar area black with slight white dust and a moderate, median, ferruginous stripe ; bristles normal ; *legs* : ferruginous ; femora, front pair proximal half, mid pair proximal two-thirds yellow, hind all yellow ; tibiae light ferruginous, tarsi yellowish, darkening outwardly ; *wing* (as Bezzi's figure 1915), costal stripe not reaching fourth vein in female, but faintly in males, hyaline part of wing faintly infuscated, more strongly towards hind margin ; third vein finely setose on its whole length ; upper cross-vein rather nearer middle of discal cell, the outer section of fourth vein over discal cell being 0·7 the inner ; point of anal cell 2·55 rest of sixth vein in male, 1·66 in female. *Abdomen :* ♀ : blackish, more or less mottled ferruginous, the yellow fascia on hind part of second segment as weak, yellowish spots on either side ; a yellowish spot on middle of fourth tergum and a pair of yellow, depressed areas on fifth separated by a ferruginous stripe ; oviscape 2·1 mm., 0·24 wing-length and 0·48 preabdomen ; legging-shaped or conical, ferruginous, mid joint yellowish, aculeus ferruginous ; sixth tergum hidden, sterna ferruginous, the fifth large. ♂ : ferruginous, hind two-thirds of second tergum yellow except at sides, a narrow, ferruginous, median stripe divides an ill-defined, yellowish, median stripe and the pair of yellow areas on fifth tergum. In the smaller male the tergum is completely blackish-ferruginous, probably through discoloration ; venter : membranes yellowish, sterna ferruginous ; terminalia light ferruginous, not dissected.

It is evident from the references made by authors that there has been some uncertainty about Fabricius' species. Bezzi, however, correctly identified a female in 1915, but as he used an unsuitable character, the yellow humeri, in his tables (1915 and 1924), it seems that later authors were misled and took specimens of *Dacus bivittatus* with yellow humeri for *D. armatus*, the yellow of the humeri being variable in *D. bivittatus*.

In 1935, having seen Collart's statement (1935 Bull. Mus. Hist. nat. Belgique, **11** p. 28—see under *D. bivittatus*) I wrote to Sir Guy Marshall, then Director of the Imperial Institute of Entomology, about the two species and he kindly sent me on loan a pair of *D. armatus* for study, the female being that identified by Bezzi in 1915. Later, I received from Dr. Hesse a copy of Fabricius' description.

It may now be noted that one important point that has been overlooked is that the hind femora in *D. armatus* are quite yellow, while in *D. bivittatus* they are ferruginous on the outer third. However, a perusal of Fabricius' description raised another query since it states " Brustseiten mit tiefer gefärbter und weisslich schimmernder Binde ", and does not mention the yellow mesopleural stripe—Sack (1908 p. 5) in his tables assumes that it is absent. Speiser (1910 p. 183) in his tables falls into another error as he says " Scutellum ohne Gelb ", because Fabricius states it is " rötlich ", but is obviously so through the discoloration that so often happens to the yellow thoracic markings in the DACINAE.

Another statement that has caused misunderstanding is that in his description of 1915 Bezzi says—in regard to the specimen he saw—" ovipositor very long and pointed ". This is correct as the ovipositor happens to be almost fully extended, but it is really not so very long compared with some other species. It seems to have misled Hendel when he described his *Dacus rubiginosus* (see under *D. bivittatus* later) as doubtless he visualised something like the oviscape of *Dacus* (*Leptoxyda*) *longistylus* Wied. and may have assumed that by " ovipositor " was meant the base (oviscape) for which the term ovipositor has often been erroneously used.

Other work and, later, the outbreak of war and events in Europe made it impossible to make enquiries about the type of *Dacus armatus* in Copenhagen, and it is only this year (1946) that a statement was received from Dr. S. L. Tuxen of the Zoological Museum at Copenhagen, fully confirming the correctness of Bezzi's identification in 1915.

Distribution.

Most references are merely records in lists of species or in tables more or less correctly interpreted from the description. A male recorded by Bezzi (1924 p. 10) from the Congo is probably the form *lulongaensis*; Perkins (1937 p. 52) mentions specimens he got from Curran, but these are probably *cucumarius*. Specimens of *D. armatus* are thus apparently scarce, and Bezzi's record of the female from Southern Nigeria seems to be the only authentic record since the original description in 1805. The type is from Guinea; Froggatt 1909 incorrectly gives *New* Guinea. I have a male, Gold Coast, Aburi, 23.iii.1911, L. Armstrong (Imperial Institute of Entomology), and another, Gold Coast, Osiem, near Tafo, 11.i.43, H. E. Box.

The biology of *Dacus armatus* is not known.

Dacus (Dacus) armatus lulongaensis Collart.

Dacus (*Dacus*) *lulongaensis* Collart, 1935, Bull. Mus. Hist. nat. Belgique, **11**, no. 1 p. 20, ♂, fig. 6. Type in Musée du Congo.

Tridacus armatus Fabr., Bezzi, 1924, Rev. zool. afr., **12**, p. 10.

On comparing a pair from Nyasaland with the description, it seems best to place *lulongaensis* as a subspecies of *armatus*. The chief difference is that the hypopleural spots are definitely separated, while the brown costal border on the wing extends more strongly to the fourth vein. There is not sufficient material to study the male terminalia.

A comparison of the female with Collart's description leaves no doubt that it is the same species or form. A few colour and other notes may be made: Length 8·8 mm., wing 8·5 mm. On the frons is a strong, brown, median spot and three lateral spots, while the lunule is also brown; the facial spots are more elongate and pear-shaped. On the thorax the yellow spot on the suture is rather widely separated from the yellow notopleural callus, and the mesopleural stripe is yellow. The wing-pattern is like that shown in Collart's figure and the proportion of the outer to the inner sections of the fourth vein over the discal cell is 0·7. Collart does not mention the legs, but in the table states that the hind femora are yellow; in this female the legs are ferruginous, the fore femora yellow on basal half, mid on basal two-fifths, and the hind all yellow; the first tarsal joint is yellowish, the hind pair most so. The abdomen is blackish ferruginous with ill-defined, yellowish patches across the hind part of the second tergum and on the middle of the third and fourth, on the fifth is a pair of large, oval, yellow areas separated by a blackish, median stripe. The conical ovipositor is chestnut with pale pubescence, length 1·8 mm., about 0·2 wing-length and 0·47 preabdomen; middle joint and aculeus coloured as base.

The male also agrees with the description, but differs particularly in the wing-pattern. The thorax is more ferruginous with only slight blackening on pleura and dorsum and the yellowish median stripe on the abdomen is not marked. On the wing, the proportion of the outer to the inner sections of the fourth vein over the discal cell is smaller, 0·63. From the figures given by Collart, in his specimen it is 0·75, and he gives this as a difference from *D. bivittatus* in a specimen of which his figures give 0·42. Such comparative measurements are, however, apt to be unreliable, and similar measurements made on only nine random specimens of *D. bivittatus* give a ratio ranging from 0·45 to 0·6. The most that can be said is that the upper cross-vein tends to be nearer the middle of the discal cell in *armatus*, farther out in *bivittatus*. The difference in the wing-pattern is perhaps just sufficient to suggest that this male may be a different species: there is only a trace of infuscation in the upper, outer corner of the first basal cell, and it extends over the fourth vein into the first posterior cell for a distance equal to about twice the height of the arc of the fourth vein.

Distribution.

The type is from the Congo. The two Nyasaland specimens are :—♂, Katambwe, Busiro, 29.ix.29, G. H. E. H(opkins)—from H. Hargreaves, and ♀, Cholo, x.1919, R. C. Wood (Imperial Institute of Entomology).

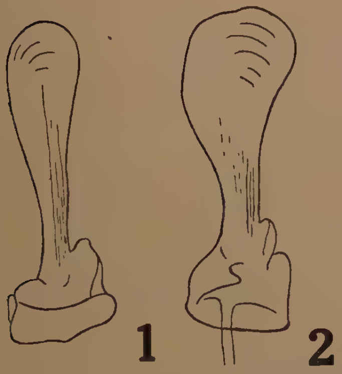
Dacus (Dacus) bivittatus (Big).

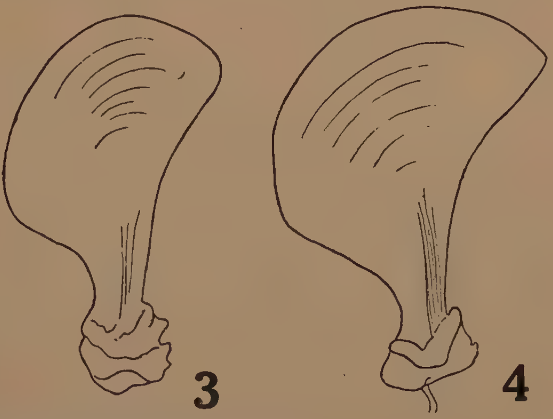
In considering this species, two points in particular may be noted. One that the name *Dacus pectoralis* Walker 1861 cannot be used as it is a homonym of *Dacus pectoralis* Walker 1859, an Oriental species. The other that it has been confused with *Dacus armatus* Fabr.

While examining numerous specimens in order to draw up the following description, it became evident that details of colour may vary very much according to the condition of the specimen, all too often teneral in reared material, or in which the yellow thoracic markings have become discoloured and reddish.

The general colour is ferruginous to chestnut (" reddish "), but most specimens available have been reared or taken in bait traps; those taken in the field tend to be blacker, and a few are distinctly black.

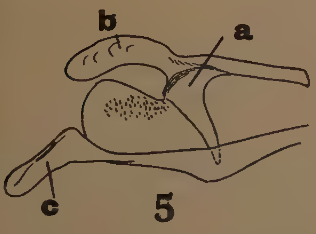
Length, ♂, 8·0 to 8·5 mm., wing 7·25 to 7·5 mm., ♀, 10·7 mm., wing 7·8 mm.—a small female length 8·25 mm. *Head:* normal, posteriorly ferruginous with a wide, yellow, postocular margin, bristles normal; *frons*, width 0·64 length, 0·33 width of head, sides more or less yellow with three brown, subintegumentary spots, a flat, ferruginous tubercle in middle, brownish ferruginous around black ocellar dot, but colour variable; *lunule* short, ferruginous; *antennae* ferruginous, third joint blackened especially on outer side, arista, base brown, flagellum black, bare; *face* yellowish brown, the two spots large, rounded to semicircular, the inner point, particularly in female, tending to be produced towards side of epistome; parafacials





Figs. 1–2.—*Dacus bivittatus* (Big.) ejaculatory apodeme.
Figs. 3–4.—*Dacus bivittatus cucumarius* Sack, ejaculatory apodeme.

narrow, genae about as wide as third antennal joint, a slight brown spot below eye, bristle small. *Thorax:* generally more ferruginous and less black, pubescence fine, pale, gold-shining; humeri yellow to almost ferruginous as will be noted, postsutural yellow stripes moderate, as long as, but rather narrower than in, *armatus*, in a few specimens there is a hint of a pale ferruginous darkening in the groove between the two hypopleural spots; this apparently increases with the melanism, and in the very black specimens recorded later as a new form, the spots are distinctly separated; bristles normal; *legs* ferruginous, fore femora yellow on proximal fourth, mid on proximal two-fifths and hind on proximal three-fifths, first tarsal joint yellow; squamae brownish, narrow; halteres yellowish brown; wing similar to that of *armatus*, anal stripe strong, costal border to middle of first posterior cell (*bivittatus*) or to fourth vein or even slightly over (*cucumarius*), but variable according to condition of specimen, rest of wing clear hyaline; point of anal cell in male 2·5, in female 1·7 rest of sixth vein. *Abdomen:* ferruginous, yellow fascia on second tergum well-marked, no marked median, yellowish stripe, but a slight narrow, median, darker



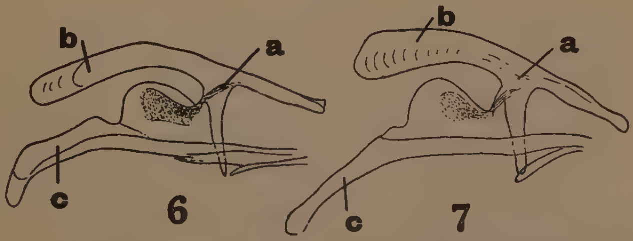
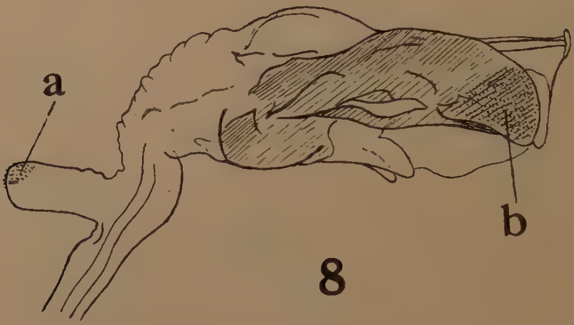


Fig. 5.—*Dacus bivittatus* (Big.), *a*, fultella; *b*, phallic apodeme; *c*, sternal apodeme.
Figs. 6–7.—*Dacus bivittatus cucumarius* Sack, *a*, fultella; *b*, phallic apodeme; *c*, sternal apodeme.

stripe, the oval depressed areas on fifth tergum not strongly differentiated ; pubescence somewhat golden yellow ; oviscape short, length 1·5 mm., about 0·2 wing-length, 0·4 preabdomen, conical or legging-shaped, ferruginous ; spermatheca (fig. 9). Male terminalia : ejaculatory apodeme (figs. 1–4) ; fultella (figs. 5–7). In the aedeagus (fig. 8) the basal sac " a " is finely setose at the end, and the large apical sac " b " is lined with close-set, blunt setae on irregular, hexagonal bases.



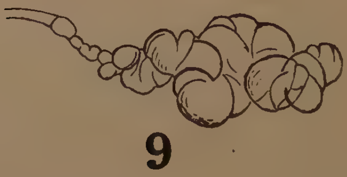


Fig. 8.—*Dacus bivittatus* (Big.), aedeagus, *a*, basal sac ; *b* apical sac.
Fig. 9.—*Dacus bivitattus cucumarius* Sack, spermatheca.

Of the various factors that have caused confusion with *armatus*, the first is the colour of the humeri. It is unfortunate that Bezzi, in his tables, used the yellow coloration of the humeri to separate *armatus* from *bivittatus*, neglecting the good character of the colour of the hind femora, although he had noted it. This seems to have led later workers, who apparently were satisfied to use his tables alone, to confuse the two. When series of *bivittatus* are examined, from any locality and even in one rearing (probably the progeny of one female), specimens are found in which the humeri may have a single anterior yellow spot, one fore and aft, a broad median stripe, may be mottled or even entirely yellow. When studying the wing-pattern, only well-coloured specimens are of any value. In West African specimens, of which only a few are available, the lower half of the first posterior cell is hyaline, or with the barest trace of infuscation, while in Central, East and South African specimens, the brown of the costal border reaches the fourth vein or just very slightly over it ; this is, on the whole, strongest in South African specimens, those from Congo and East Africa tend to have the lower part of the first posterior cell paler. Bezzi and Collart use this difference to separate *bivittatus* and " *pectoralis*," but do not seem to attach any regional significance to it.

Finally, from an examination of the male terminalia* a reduction in size in some structures was noted in the West African specimens as compared with those from other parts of Africa.

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*A detailed study of the terminalia of some male Trypetidae has been made by the author in Memoir No. 1, 1947, of the Entomological Society of Southern Africa.

1. Ejaculatory apodeme: in Nigeria and Sierra Leone specimens narrow (figs. 1, 2), while in specimens from Kenya, Tanganyika, Congo, Southern Rhodesia and Natal, it is wide and fan-shaped (figs. 3, 4).

2. Phallic apodeme: in the West African specimens examined (fig. 5) it is about 0·5, and in those from Kenya, etc. (figs. 6, 7), about 0·66 the length of the fultella.†

3. Sternal apodeme: in West African specimens about 0·3; in others, about 0·5, occasionally varying to 0·4 the length of the fultella.

4. No differences were apparent in the aedeagus (fig. 8), but this complicated structure cannot be considered in detail here.

For a complete study of the species more material is needed from West Africa, particularly the area from Kamerun to Angola. The wing-pattern difference occurs in both sexes, but otherwise the females show no appreciable differences, not even in the spermatheca (fig. 9). However, in view of the differences in the male terminalia, it seems best to recognise two subspecies to be identified partly on wing-pattern and male terminalia, partly on locality. The typical form, from the West Coast, is *bivittatus* (Big.), while to the other the name *cucumarius* Sack must be applied. An analysis of the bibliography and synonymy follows.

Dacus (Dacus) bivittatus bivittatus (Bigot).

Leptoxys bivittatus Bigot, 1858, Arch. Ent. (Thomson), **2** p. 374, pl. 10, fig. 7, ♀. Type in coll. Thomson. Bagun.

Dacus bivittatus (Big.) Bezzi, 1908, Boll. Soc. ent. ital., **39** pp. 138, 147; 1908, Boll. Lab. Zool. Portici, **3** pp. 292, 297. Speiser, 1910, Wiss. Z. Exp. Kilim.-Meru, **10** p. 183. Enderlein, 1911, Zool. Jahrb., Abt. Syst., **31** p. 409. Munro, 1933, Amer. Mus. Novit., no. 597 p. 3, the Liberia specimen. Perkins, 1937, Proc. roy. Soc. Qd, **48** p. 52.

Tridacus bivittatus (Big.), Bezzi, 1915, Bull. ent. Res., **6** pp. 89, 92, fig. 6; 1924, *ibid.* **15** p. 82.

Dacus cucumarius Sack, Silvestri, 1913, R. Accad. Lincei, (5) **22** pp. 205–206, parasite, Kamerun.

Dacus armatus Bez. *nec* Fabr. Bezzi in Silv., 1913, Boll. Lab. Zool. Portici, **8** p. 89, fig. xxiii; 1914, Bull. Div. Ent. Hawaii no. 3 p. 89, pl. viii. fig. xxiii; Bezzi, 1914, Boll. Lab. Zool. Portici, **8** p. 302; 1915, Bull. ent. Res., **6** p. 92—synonymy.

Dacus bipartitus Graham, 1909, Rep. Ent. Obs. S. & Centr. Ashanti, 1907, p. 11, tab. 4, figs. 7–8, ♂♀. Type in British Museum; 1910, Bull. ent. Res., **1** p. 167, pl. xiii, figs. 1–2. Bezzi, 1909, Boll. Lab. Zool. Portici, **3** pp. 292, 297; 1914, *ibid.* p. 302. Patterson, 1913, Rep. Ent. Gold Coast agric. Dept., **1913** pp. 18–27.

Note: *Dacus bipartitus* Bez. 1914 (*nec* Grah.) in Silv. Bull. Div. Ent. Hawaii no. 3, p. 90, is not this species but *D. momordicae* Bez.

Distribution.

The typical form is recorded from Sierra Leone, Liberia, Ashanti, Gold Coast, Nigeria, Kamerun and the Island of Principe. It has been reared from cucumber and from cucurbits generally, as well as *Cucumis* and *Momordica*, the last two presumably wild hosts.

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† *Fultella,* a taxonomic term applied to the modified framework supporting the phallus in Trypetidae: see Memoir just quoted.

Dacus (Dacus) bivittatus cucumarius Sack.

Dacus cucumarius Sack, 1908, Ber. senckenb. naturf. Ges., **1908** p. 10, fig. Type in Mus. Senckenberg, Frankfurt. Bezzi, 1908, Boll. Lab. Zool. Portici, **3** pp. 292, 298. Speiser, 1910, Wiss. Z. Exp. Kilim.-Meru, **10** p. 183.

Tridacus cucumarius (Sack) Bezzi, 1915, Bull. ent. Res., **6** p. 92.

Dacus bivittatus (Big.) Mason, 1916, Rep. Govt. Ent. Dep. Agr. Nyasaland, **1916** pp. 19–22. Morstatt, 1913, Der Pflanzer, **9** pp. 288–296. Munro, 1933, Amer. Mus. Novit., no. 597, p. 3.

Tridacus bivittatus (Big.) Bezzi 1924, Rev. zool. afr., **12** p. 10. Munro, 1925, Ent. Mem. Un. S. Afr. Dep. Agr., **3** p. 41.

Dacus pectoralis Walker, 1861, Trans. ent. Soc. Lond., (N.S.) **5** p. 322, type in British Museum—homonym of *Dacus pectoralis* Walker, 1859, J. Proc. Linn. Soc. Lond., Zool., **3** p. 114, oriental species. Bezzi, 1908, Boll. Soc. ent. ital., **39** pp. 139, 146; 1908, Boll. Lab. Zool. Portici, **3** pp. 292, 296. Froggatt, 1909, Rep. paras. inj. Ins., p. 99. Speiser, 1910, Wiss. Z. Exp. Kilim.-Meru, **10** p. 183. Enderlein, 1911, Zool. Jahrb., Abt. Syst. **31** p. 409. Bishop, 1934, Fmg S. Afr., reprint no. 71. Munro, 1935, Ent. Mem. Un. S. Afr. Dep. Agr., **9** p. 18. Perkins, 1937, Proc. roy. Soc. Qd, **48** p. 52.

Tridacus pectoralis (Walk.) Bezzi, 1915, Bull. ent. Res., **6** p. 92; 1924, *ibid.*, **15** p. 82; 1924, Ann. S. Afr. Mus., **19** p. 455, pl. xii, fig. 2; 1928, Ann. Transv. Mus., **12** p. 330. Munro, 1925, Ent. Mem. Un. S. Afr. Dep. Agr., **3**, p. 41; 1926 *ibid.* **5** p. 19; 1929, Ann. S. Afr. Mus., **29** p. 2. Bianchi & Krauss, 1937, Hawaii. Planters Rec., **41** pp. 299–306.

Dacus armatus Curran *nec* Fabr. Curran, 1927, Bull. Amer. Mus. nat. Hist., **57** p. 88.

Dacus (*Dacus*) *armatus* Collart *nec* Fabr. Collart, 1935, Bull. Mus. Hist. nat. Belgique, **11** p. 28 and 1940, *ibid.* **16** p. 2, including as varieties *bivittatus* Big., *pectoralis* Walk., and *hulstaerti* Coll.

Dacus (*Dacus*) *rubiginosus* Hendel, 1928, Ent. Mitt., **17** p. 347, type in Deutsche Ent. Mus., Berlin-Dahlem.

Following what has been said, there is no doubt that *cucumarius* Sack and *pectoralis* Walk. 1861 are the same. That Walker should have given the same name to two species within a short time is curious. It seems, however, that the paper in which he described the African species was read on 2nd February, 1857, but was not published till 1861; in the meantime, not having a printed copy of the paper, nor perhaps even a manuscript copy, he overlooked it, and gave one of the names in it to an oriental species in 1859. As interpreted by Collart in his tables and descriptions, it is evident that his *armatus* is *cucumarius*, since he did not recognise the true *armatus*. Further, his var. *hulstaerti* is based on a discoloured specimen of *cucumarius* in which the yellow markings have become reddish—he mentions another specimen in which only the scutellum is reddish. As regards *rubiginosus*, it would seem that as the type specimen has yellow humeri, Hendel considered it could not be either *bivittatus* or *pectoralis* according to Bezzi's tables. Then, as it did not have a " long ovipositor " it could neither be *armatus*. It may be noted that Hendel says the base of the ovipositor in *rubiginosus* is three-fifths the length of the fifth tergum, but this measurement seems to refer merely to the part projecting behind! He made a similar mistake about his *D. furcatus* (=*D. punctatifrons* Ksh.) described at the same time, a fact that has been checked on one of his paratypes. As far as may be judged, the material concerned was all in very poor condition, either very teneral or badly preserved in alcohol and dried without suitable precautions. His description agrees sufficiently for his species to be regarded as a synonym of *cucumarius* except in one point: he says the hind femora are ferruginous on the distal half, not third as usual, but this may be due to the condition of the specimens.

It may be noted that all specimens identified by me since 1926 as either *bivittatus* or *pectoralis* are *cucumarius* except a few from Sierra Leone and Nigeria which should be *bivittatus.*

Distribution.

In South Africa the fly has been recorded from Retreat (near Capetown), along the coastal area from Great Brak River and Mossel Bay to Port Elizabeth, thence to East London, Komgha and Port St. John, Fort Beaufort being the most inland locality. In Natal it is recorded from Port Shepstone in the south to Durban and Eshowe, Cedara being the most inland locality. In the Transvaal in the east from Barberton, Kaapmuiden and Nelspruit, in the north from the Njelele River, and centrally from Rosslyn and Pretoria. There is one record from South West Africa. It has not yet been found in the Karoo and highveld areas generally. Beyond the Union it is recorded from various localities in Portuguese East Africa, Southern Rhodesia, Congo, Tanganyika, Nyasaland, Uganda, Kenya, Zanzibar and Eritrea.

The insect is probably more or less common in the areas indicated, but local distribution may be discontinuous. It is undoubtedly a tropical and subtropical species. It may have been more restricted originally, but may be expected to extend its range as far as it can find conditions suitable. It is felt that the record from Retreat in 1941 is a recent incursion, but such date records need not carry too much weight. The earliest South African records are: 1904, Komgha; 1906, Great Brak River and 1913, Mossel Bay, but the first record from Port Elizabeth is 1926. It is interesting to note that it was first noted in Pretoria in 1941, but had been taken at Rosslyn, some ten miles north-west of Pretoria, in 1931. Pretoria is, however, to the south of the Magaliesberg and is distinctly more temperate in climate than Rosslyn to the north of the mountains where there is a narrow, subtropical zone along the northern slopes of the range. Further, in Pretoria, the flies did not put in an appearance till well towards mid-summer. This late appearance is a phenomenon also noticed in the case of *Ceratitis capitata* Wied., especially in parts of the Orange Free State, but which cannot be discussed here.

Food-plants.

Records of food-plants have been taken from data on labels attached to specimens and from available publications, that of Bianchi and Krauss, dealing with the search for fruit-fly parasites in East Africa, being of interest.

The fly typically infests cucurbitaceous fruits but five other kinds are recorded—the cultivated fruits pawpaw, tomato, both regularly and severely, and granadilla, and Bianchi and Krauss record *Vitex* sp. and *Dioscorea macroura* from East Africa.

Food-plants recorded in South Africa are:

Wild: *Cucumis metuliferus, Coccinea palmata, Momordica involucrata, Peponium McKennii, Peponium Vogelii* and *Sphaerosicyos sphaericus* (all Cucurbitaceae).

Cultivated: Pumpkin, Sweetmelon (spaanspek), Kaffir melon, Calabash, Pawpaw (*Carica papaya*), and tomato.

From other parts of Africa records are less numerous.

Uganda: Tomato, squash, *Cucurbita* sp., *Vitex* sp.

Kenya: Marrow, melon and an unidentified Cucurbit.

Zanzibar: *Luffa aegyptica, Momordica charantia* and cucumber.

Nyasaland: Cucumber.

Tanganyika: Pumpkin, chou-chou (*Sechium edule*), *Peponium* sp., *Luffa* sp., a cucurbit sp., granadilla (*Passiflora quadrangularis*) and *Dioscorea macroura.*

Southern Rhodesia: Melon, cucumber, marrow.

General Biology and Habits.

Little, if anything, is known of the biology of this species. Under favourable, that is, more tropical, conditions, it is present all the year round attacking what fruits may be available. At Durban Mr. Marriott has noted that if pumpkin plants are not cleared from the lands, they can survive till the second year and so provide an early breeding ground for the fly. In areas where frost occurs, pumpkin plants are killed off during the winter, but *cucumarius* is not usually found in such places.

The fly infests the fruits and may attack them at any stage of their growth; in one instance Mr. Marriott found larvae in the flowers of *Sphaerosicyos sphaericus.* In Pretoria it was noticed that only older pumpkins in which the rind had already hardened were attacked, but this may have been due to the late arrival of the fly.

The period from egg to adult has not been recorded in detail, but may be from one to two months. In an infestation of pawpaws, the larvae were full-grown in 19 days after egg-laying, and flies emerged 30 days after pupation. Bishop (1934) dealing with *D. ciliatus* Lw., *D. vertebratus* Bez. and *D.* " *pectoralis* " together, gives 3 to 5 days for the hatching of the eggs, and up to 14 days for the pupal period, but does not mention the larval period. The number of generations a year has not been observed, but it is thought there may be two or three under favourable conditions, but perhaps only one if the fly is restricted by food supplies.

Adult flies probably live for some time, over-wintering as adults when necessary. They feed on honey-dew and other sweet exudations available. On a specimen taken by C. F. M. Swynnerton at Mount Chirinda, Southern Rhodesia, the label states "frequents ripe bananas," and the collector notes that the fly is a mimic of the Eumenid wasp *Odynerus vulneratus* Sauss.

Common Name.

Dacus bivittatus (Big.) may be referred to as the larger, common pumpkin or cucurbit fly. I have used the term " pectoral pumpkin fly " and this could be retained as a specific common name, preserving at the same time a reference to Walker's name.

Parasites.

Dacus bivittatus does not appear to be parasitised to any extent. Silvestri records *Tetrastichus giffardi* Silv. from puparia from Kamerun, while Bianchi and Krauss record only two specimens of parasites from Zanzibar, one a *Dirhinus* sp., the other *Spalangia afra* Silv.

Dacus (Dacus) bivittatus cucumarius Sack., form **atrescens nov.**

Under this name are recorded some specimens that may be taken to represent a melanic variation, or perhaps phase, of *cucumarius.* The males are strongly black and the hypopleural spots separated by a narrow, ferruginous stripe, in one rather broadly so; the females are not quite so black as the males. Otherwise the specimens agree with the more typical *cucumarius*; in the few specimens available there is an indication of the same variation in the yellow colour of the humeri and the costal border on the wing reaches the fourth vein although rather pale in the lower part of the first posterior cell which in one male is practically colourless. The male terminalia, like those of *cucumarius*, have a wide, fan-shaped ejaculatory apodeme and a long phallic apodeme.

Holotype ♂, allotype ♀, paratypes 1♂, 1♀, Kenya, Bwamba, 7.viii.1946, V. G. L. van Someren. Paratypes, 2♂♂, Belgian Congo, Tshibinda, 21–27.viii.1931, Miss A. Mackie (Imperial Institute of Entomology).

Dacus theophrastus Hering.

Hering, 1941, Siruna Seva, No. 3, p. 5, fig. 3.

Since this was described as a " form " of *Dacus armatus*, Fabr., it is mentioned here. In the colouration of the legs and in wing-pattern it has nothing to do with *D. armatus* and seems to represent a distinct species.

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